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# PLANTERS' RECORD

VOL. XLIII



**A quarterly paper devoted to the sugar interests of Hawaii,  
and issued by the Experiment Station for circulation among  
the plantations of the Hawaiian Sugar Planters' Association.**

JANUARY

TO

DECEMBER

# THE HAWAIIAN PLANTERS' RECORD

VOL. XLIII

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ORGAN OF THE EXPERIMENT STATION OF THE  
HAWAIIAN SUGAR PLANTERS' ASSOCIATION

HONOLULU

1939

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ILLUSTRATIONS APPEARING ON THE COVERS OF  
VOLUME XLIII

FIRST QUARTER



Delays in weed control were responsible for these differences in cane growth.

## SECOND QUARTER



The flower of the Baobab tree (*Adansonia digitata*), an African species of which there are at least two individuals of fruiting age in Honolulu. There are many of these trees on the plains of Uganda which will exceed both in age and in diameter of trunk the famed redwoods of California.

## THIRD QUARTER



The greatly increased internode elongation of the "C" over the "A" stalks was an effect of intermittently reduced periods of direct sunlight during the "boom" stage of growth.



## FOURTH QUARTER



FIRST RATOON OF ORIGINAL STOOL OF 31-1389, FIELD 17,  
MAKIKI PLOTS, MARCH 18, 1932

From this stool 31-1389 was extended to over sixteen thousand acres within eight years.





# THE HAWAIIAN PLANTERS' RECORD

Vol. XLIII

FIRST QUARTER 1939

No. 1

*A quarterly paper devoted to the sugar interests of Hawaii and issued by the Experiment Station for circulation among the plantations of the Hawaiian Sugar Planters' Association.*

## **In This Issue:**

### *A Simple Apparatus for the Rapid Determination of Moisture by the Carbide Method:*

A rapid and extremely simple method is described for the determination of soil moisture. The method eliminates drying of the specimen in a heated oven. With a slight additional simplification it may be adapted for rough estimations of soil moisture in the field by using a measuring device to gauge a preselected volume of sample instead of weighing it on a laboratory balance.

The method as described requires but 10 minutes to perform a soil moisture determination.

### *Studies in Experimental Technique:*

The value of laying out field experiments so that their results may be studied by modern refinements of statistical methods is clearly shown in the actual interpretation of the results from a potash experiment.

### *A Pictorial Showing the Effects of Delayed Weed Control Upon Subsequent Growth of Sugar Cane:*

A series of pictures tells a graphic story of the effect of delayed weeding upon the cane which is growing in soils having specific variations in available plant foods.

### *Mineralizable Nitrogen in Some Hawaiian Soils:*

Ninety per cent or more of the reserves of organic soil nitrogen (total nitrogen) are not available to plant life until after "mineralization" takes place. The term "mineralization" implies naturally occurring microbiological changes in the soil in which soil organic nitrogen is transformed to compounds of ammonia and to

nitrates, both of which are capable of immediate utilization by a growing crop. The authors present discussion and data in plain terms supporting the premise that organic soil nitrogen is readily mineralizable under favorable conditions. Their laboratory determination of this property is described. (It is adaptable to R. C. M.) Definitions of terms are given fully. The authors point out the importance and significance of the *rates* and of the *phases* at which mineralization occur and, more importantly, they deal with the relationships they found to exist between mineralizable soil nitrogen and the absorption of this nutrient by indicator plants in a controlled culture study.

---

#### *Plant Food Ratios for Sugar Cane Fertilizers:*

Various ratios of N:P:K in a complete fertilizer mixture, which were tested on three different soil types, have given little encouragement that an optimum ratio may be specified. The sugar yields secured were not significantly different even when some rather wide differences in N:P:K ratios were used, and those ratios which were associated with the "best" yields were not in general agreement with the known requirements for the soils studied.

---

#### *Colloids in the Sugar Mill:*

The colloidal impurities present in cane juice are of many different kinds. For each type, a discussion is given as to the sources and properties of the compounds, the changes which take place during the passage of the juice through the sugar mill, and the effects of the substances on the work of the mill.

---

#### *Availability of Insoluble Phosphates:*

An experiment is described in which it was shown that raw rock and reverted phosphates may be equally as available to sugar cane as calcium superphosphate on acid, high phosphate-fixing soils. Raw rock phosphate was found to be more resistant to fixation by the soil than either of the other forms studied.

---

#### *Sixth Congress of International Society of Sugar Cane Technologists:*

A general report on the Louisiana sugar industry and the papers presented at the gathering of delegates are presented by the chairman of the delegation from Hawaii. Both the report to the Experiment Station and the report to the Hawaiian Sugar Technologists were presented by the writer at the annual meetings of the Hawaiian Sugar Planters' Association and the Hawaiian Sugar Technologists in 1938.

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# A Simple Apparatus for the Rapid Determination of Moisture by the Carbide Method\*

By EDWARD T. FUKUNAGA and L. A. DEAN†  
Hawaii Agricultural Experiment Station

In many agricultural practices it is desirable to have available a simple and rapid method for the determination of moisture. This communication describes a simple apparatus for such a purpose, utilizing the calcium-carbide method.

Calcium carbide reacts with water to form acetylene as indicated below:



Thus it follows that 1 gram of water will react with an excess of calcium carbide to produce 690 c.c. of acetylene at 30° C. The principle underlying the carbide method is the direct determination of the moisture of a substance by causing the moisture to undergo such a reaction and measuring the acetylene produced, either by measuring the volume at constant pressure or the pressure at constant volume. The latter procedure apparently permits the simplest and most compact form of apparatus.

## APPARATUS AND MATERIALS

Fig. 1 illustrates the apparatus found suitable for determining moisture by the calcium-carbide method. This consists of a welded cylindrical bomb, 4 inches in diameter and 7 inches in depth, made of an aluminum alloy. A machined flange and plate, held together by bolts and wing nuts, constitute the head of the bomb. The pressure-tight seal between the flange and plate is accomplished by a cork gasket shellacked to the under surface of the plate, and a coating of viscous grease which is smeared on the upper surface of the flange. Into a threaded opening in the plate is fitted a 30-pound pressure gauge with a 4-inch dial. The plate is also equipped with a Weston all-metal thermometer, the stem of which extends into the bomb for a distance of 3½ inches.

The equipment necessary to operate the bomb is:

1. A tin can 4 inches high and of such diameter that it just fits within the bomb.
2. A metal container or vial 1½ inches in diameter and 1½ inches high.
3. Commercial calcium carbide which has been crushed to pass a screen with 0.5 mm. mesh.

## DETERMINATION OF MOISTURE

The above-described apparatus may be used for determining moisture in samples of material containing between 0.5 and 3 grams of water. A sample of the material is weighed into the tin can (item 1 above). The metal cup (item 2), filled

\* Contribution of the Department of Chemistry and Soils. Published with the approval of the Director of the Hawaii Agricultural Experiment Station.

† Assistant in Chemistry and Assistant Chemist, respectively.

with calcium carbide and uncovered, is placed in an upright position in the tin can, and the can is inserted into the bomb, which is then sealed. The bomb is held in an inclined position in order to tilt over and discharge the carbide upon the specimen in which moisture is to be determined. The apparatus is then returned to an upright position and shaken with a swirling motion for about a minute. This causes an almost instant reaction between the carbide and the moisture of the sample. It is necessary to shake only until no further increase in pressure is noted. The temperature and pressure are then read and from these data the amount of acetylene generated, which is a measure of the moisture content of the sample, is calculated by applying the gas laws. In general practice, however, it has been found advantageous to construct charts from which the amount of water corresponding to a given pressure can be quickly determined.

The only necessary calibration of the apparatus is an accurate estimation of the volume of the bomb. This may be quite simply ascertained by measuring the increase in pressure caused by a known amount of water.

### DISCUSSION

The above-described procedure was applied to a number of soils, each represented by samples ranging in moisture content from maximum field capacity to below the wilting coefficient. Results were compared with determinations made by the oven-drying method, and a constant difference was found to exist between the two types of measurement. Consequently, the utility of the rapid method as an aid to irrigation control is suggested if the critical soil moisture constants of specific soil types are predetermined on the calcium-carbide basis.

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*Note by Associate Editor.*—A series of independent soil moisture determinations were made by F. Ray Van Brocklin in the chemistry laboratory of the Experiment Station, H.S.P.A., in which the Fukunaga-Dean bomb was compared with conventional oven drying of soil for the purposes of checking the bomb.

While it is recognized that several factors in oven drying of soil detract from the accuracy of this method in arriving at a true moisture value for the specimen it is also recognized that the method itself is one almost universally employed.

It is patent then, we believe, that oven drying should not be considered a criterion of accuracy in the determination of soil moisture.

We found duplicate bomb determinations to be consistent and in most cases to vary from "oven-dry" data by a margin reasonably to be expected as due to differences in the methods used and to errors in manipulation. In all cases the bomb results were lower than oven data, varying from the latter values between 3 per cent and 18 per cent.

One set of comparable data follows:

Per cent H <sub>2</sub> O by bomb	Per cent H <sub>2</sub> O by oven at 102° C.
3.74	3.86
5.05	6.21
5.70	6.20
6.80	8.14
7.98	9.59
13.10	15.29
13.90	16.31
18.30	19.22

F. E. H.

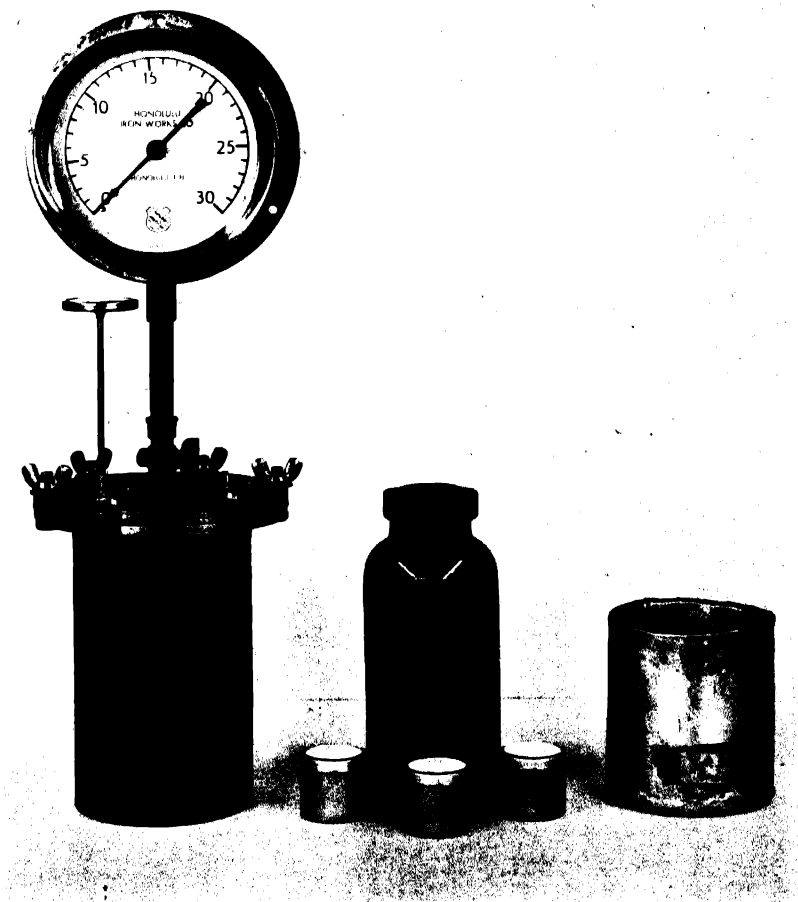


Fig. 1. Apparatus and accessories for determining moisture by the carbide method. (From left to right: bomb, metal vials  $1\frac{1}{2}$  inches in diameter by  $1\frac{1}{2}$  inches high, powdered calcium carbide, and tin can 4 inches in diameter by 4 inches high.)

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## Studies in Experimental Technique

### Selection of Layout: Blocks *versus* Latin Squares

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By R. J. BORDEN

The total amount of variation due to all causes, between the plot yields of a field experiment, can be divided into several relevant portions, and various statistical measures can be used to assess these portions of the total variation to their respective influences. The most generally used measure today is that afforded by Fisher's analysis of variance.

The total sum of the squares of deviations of each plot yield from the mean yield *of all plots* included in the experiment gives a measure of the total amount of variation between the different plots. The sum of the squares of deviations of each plot yield from the mean yield of those *plots with the same treatment* gives a measure of the discrepancies *within* the treatments, which may be due to the position of the plots on the test area, to differences in the interactions of treatment and its position, as well as to the unknown, indeterminable effects upon the various plot yields. The difference between this total sum of squares and the sum squares within the several treatments is the amount of variation between the treatments which has been contributed, and hence is due to the treatments that were given.

Corresponding to these sums of squares (within treatments and between treatments) into which the total sum squares can be divided, we have their mean squares which are calculated by dividing each by its respective number of degrees of freedom. Thereafter the ratio between the mean square for treatment and the mean square for the unknown effect or "error," affords us our test of the significance of the yield differences between the treatments.

As indicated above, the amount of variation *within* the treatments may be due to several causes. When one or more of these can be ascertained and the amount of variation which is contributed thereby can be determined, such an amount can no longer be considered part of the unknown variance or "error" of the experiment which is used in the ratio with the treatment variance for the test of significance. Hence properly designed experiments will be installed in such a way that much of the positional variance and possibly some of the interaction effects can be calculated and used to reduce the unknown or residual error, thus allowing for a more reliable interpretation of the effect of treatment.

We have a good example for this discussion in data taken from the harvesting results of Hamakua Mill Company's Experiment 41 AK, 1938 crop. In this field test, the individual plots were .05 acre in size, with 8 lines per plot. There were 9 replicates of 3 treatments: "G"—No  $K_2O$ ; "H"—175 pounds  $K_2O$  per acre; and "I"—350 pounds  $K_2O$  per acre. The arrangement of plots\*, with plot numbers, treatment identities, and cane yields (tons per acre) are given below; also, the

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\* This arrangement would be criticized by some statisticians because the plots have not been assigned at random.

nine "blocks" and three "squares" as used in the statistical analysis that follows have been indicated:

Block No. 1	Block No. 2	Block No. 3	Block No. 4	Block No. 5	Block No. 6	Block No. 7	Block No. 8	Block No. 9
55 G 53.1	56 H 62.6	57 I 51.0	58 G 43.3	59 H 44.0	60 I 55.7	61 G 38.3	62 H 56.8	63 I 53.5
64 H 65.3	65 I 63.1	66 G 53.9	67 H 36.3	68 I 45.5	69 G 48.5	70 H 48.5	71 I 49.5	72 G 42.9
73 I 56.6	74 G 57.2	75 H 50.1	76 I 48.9	77 G 42.8	78 H 43.1	79 I 46.9	80 G 44.9	81 H 47.9
SQUARE NO. 1			SQUARE NO. 2			SQUARE NO. 3		

When these 27 plots have been so arranged that we can calculate that part of the total variation of the experiment which was due to the difference in treatment, and also that part which was due to the position which these 9 blocks of plots occupied on the test area, so that we may get a more reliable estimate of the uncontrolled error of the experiment from which we are to determine the significance of treatment differences, we have the following arrangement of data and their analysis:

Block no.	Plot nos.			Plot yields			Block totals
	"G"	"H"	"I"	"G"	"H"	"I"	
1.....	55	64	73	53.1	65.3	56.6	175.0
2.....	74	56	65	57.2	62.6	63.1	182.9
3.....	66	75	57	53.9	50.1	51.0	155.0
4.....	58	67	76	43.3	36.3	48.9	128.5
5.....	77	59	68	42.8	44.0	45.5	132.3
6.....	69	78	60	48.5	43.1	55.7	147.3
7.....	61	70	79	38.3	48.5	46.9	133.7
8.....	80	62	71	44.9	56.8	49.5	151.2
9.....	72	81	63	42.9	47.9	53.5	144.3
Treatment totals .....				424.9	454.6	470.7	1350.2 Grand total
Averages .....				47.2	50.5	52.3	

#### ANALYSIS OF VARIANCE

Due to	Sum of squares	Degrees of freedom	Mean variance
Total .....	1398.24	26	
Treatment .....	119.96	2	59.98
Blocks .....	938.29	8	
Error .....	339.99	16	21.25

$${}^{\prime\prime}F^{\prime\prime} = \frac{59.98}{21.25} = 2.82 \quad {}^{\prime\prime}F^{\prime\prime} \text{ required for minimum significance} = 3.63.$$

From this first analysis, we are forced to conclude that such differences between the treatment averages which were secured may quite likely have been the effect of chance; in other words, a definite response to the known treatment differentials which were applied was not reliably proven, for the uncontrolled or error variance

of the experiment was too large in its relation to the treatment variance to warrant any great degree of confidence in the latter.

However, when it is possible to break down the total variation of an experiment more thoroughly than can be done by simply determining the treatment and the block variations, a still further reduction of the error variance is obtained. The triple Latin Square arrangement allows for just such a procedure, for we can secure the amounts of the total variation which has been contributed by the rows of plots and by the columns of plots in the 3 Latin Squares, as well as that contributed by the separate Latin Squares themselves, and also by the interaction of the squares and the treatments.

Using these same data, which fortunately can be studied on the basis of an arrangement of three Latin Squares, we have the following:

	Plot nos.	Total row yields	Plot nos.	Total column yields	Plot nos.	Total treatment yields "G" "H" "I"		
SQUARE NO. 1:	55,56,57	166.7	55,64,73	175.0	55,74,66	164.2		
	64,65,66	182.3	56,65,74	182.9	64,56,75		178.0	
	73,74,75	163.9	57,66,75	155.0	73,65,57			170.7
Square No. 1 total yield		512.9		512.9				
SQUARE NO. 2:	58,59,60	143.0	58,67,76	128.5	58,77,69	134.6		
	67,68,69	130.3	59,68,77	132.3	67,59,78		123.4	
	76,77,78	134.8	60,69,78	147.3	76,68,60			150.1
Square No. 2 total yield		408.1		408.1				
SQUARE NO. 3:	61,62,63	148.6	61,70,79	133.7	61,80,72	126.1		
	70,71,72	140.9	62,71,80	151.2	70,62,81		153.2	
	79,80,81	139.7	63,72,81	144.3	79,71,63			149.9
Square No. 3 total yield		429.2		429.2				
Grand total treatments: G=424.9; H=454.6; I=470.7.								

Our analysis of variance now becomes the following:

Due to	Sum of squares	Degrees of freedom	Mean variance
Total .....	1398.24	26	
Treatments .....	119.96	2	59.98
Rows (137.87+65.87+51.80)= .....	255.54	(2+2+2)=6	
Columns (65.53+27.64+15.55)= .....	108.72	(2+2+2)=6	
Squares .....	682.74	2	
Interaction=[980.11-(119.96+682.74)] .....	177.41	[8-(2+2)] 4	
(Treatments and Squares)			
Error .....	53.87	6	8.98
${}^{\text{''F''}} = \frac{59.98}{8.98} = 6.68$			
${}^{\text{''F''}} \text{ required} = 5.14$			

$$SEd = \sqrt{\frac{8.98}{9}} \times 2 = 1.41$$

From this analysis it will be seen that the total and the treatment sums of squares check with the previous analysis (1398.24 and 119.96 respectively) as they should.

do, but that the corresponding figure for positional factors (blocks) which was 938.29 has now been increased to a total of 1224.41 ( $255.54 + 108.72 + 682.74 + 177.41$ ), and that this has brought about a considerable reduction in the sum of squares due to error—from 339.99 to 53.87.

This second analysis of the total variation of the experiment has made it possible to determine the effect contributed by the positional variance with more refinement than was possible with determinations of the block variance alone; it has likewise enabled the effect of the interaction between treatment and position on the test area to be measured. Such measurable effects are legitimately deducted from the total variance, thus reducing the unknown effect or error variance, upon which the true estimate of significance is based.

Here, then, we find our original conclusion may be changed. The error variance now appears sufficiently small in relation to the treatment variance to justify our conclusion that the yield differences obtained are not likely the effect of chance, and hence are probably the effect of the known treatment (potash) differentials which were applied.

Thus the value of laying out field experiments so that their results may be studied by modern refinements of statistical methods is exemplified: without this multiple Latin Square arrangement there could have been considerable doubt that the observed yield differences were due to applications of potash; with this Latin Square arrangement and our subsequent analysis, we can feel more confident that potash was actually responsible for the yield increases that were obtained.

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# A Pictorial Showing the Effects of Delayed Weed Control Upon Subsequent Growth of Sugar Cane

By R. J. BORDEN

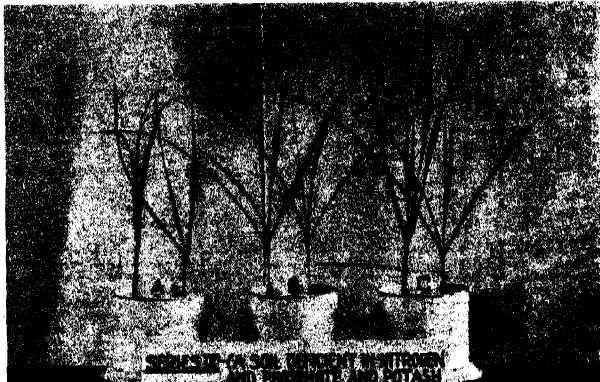


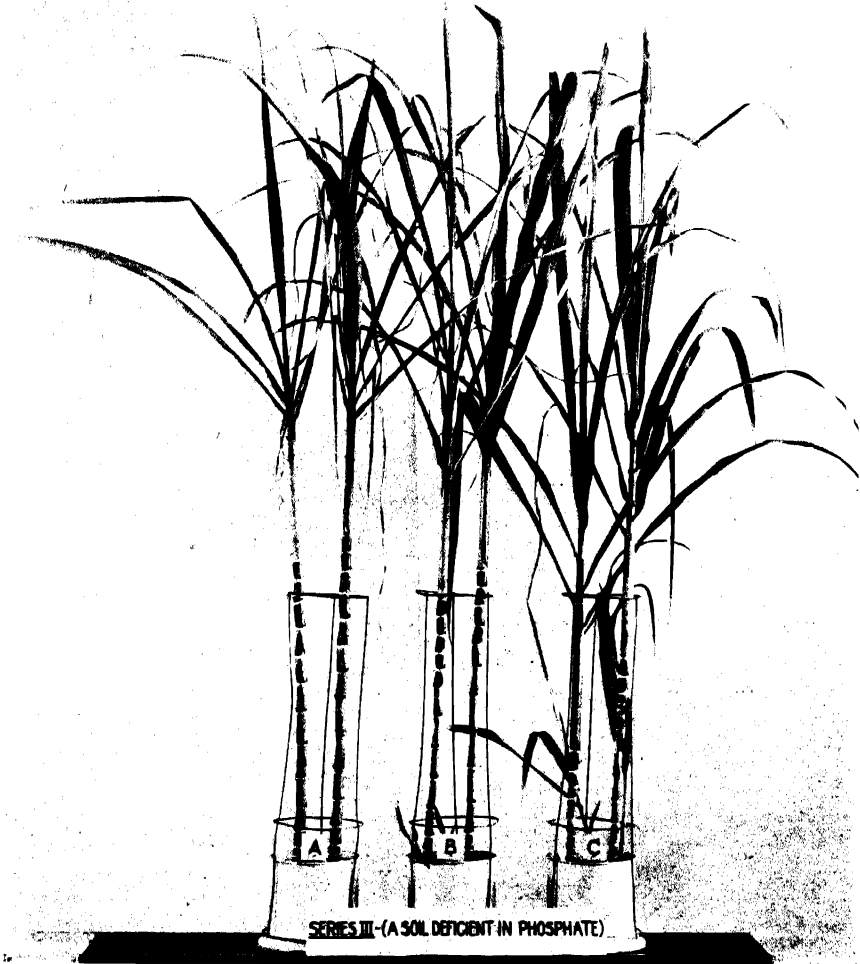
- A—Cane growth at 8 weeks; no weeds allowed to grow.  
 B—Cane growth at 8 weeks; weeded at 6 weeks.  
 C—Cane and weed growth at 8 weeks.

*NOTE.*—In all instances when pots were weeded, all weed growth was crushed and returned to the surface of the soil in the pot in which it grew. Nearly complete decomposition of this material had taken place when the cane was 4 months old; hence the plant food which had been taken up by the weeds had *presumably* been returned to the soil, and logically it should have been made available for subsequent cane growth. The probable fallacy of this presumption is indicated in the pictures which follow which were taken when the plants were 7 months old. It appears quite certain that the growth which has been lost by this crop will not be made up.

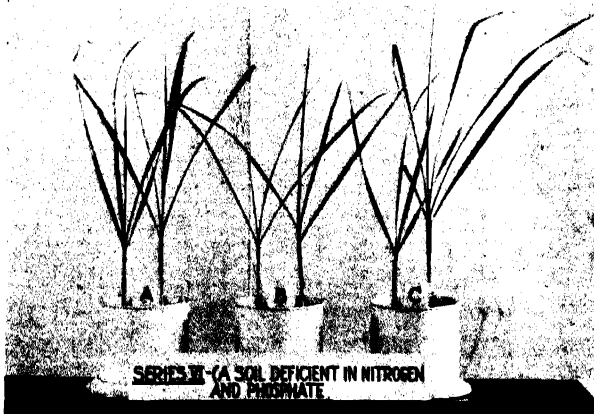


A—No weeds allowed to grow. B—Weeded at 6 weeks. C—Weeded at 8 weeks.





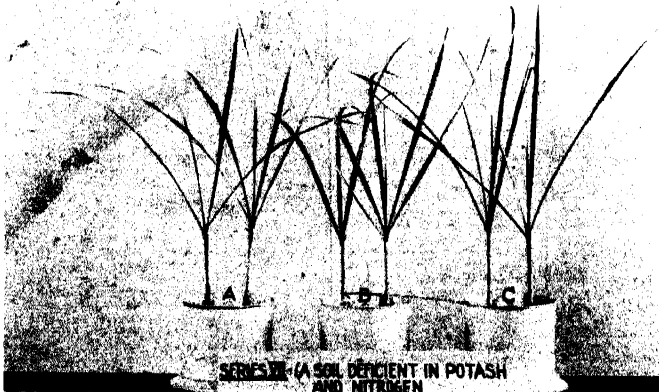
*A*—No weeds allowed to grow. *B*—Weeded at 6 weeks. *C*—Weeded at 8 weeks.





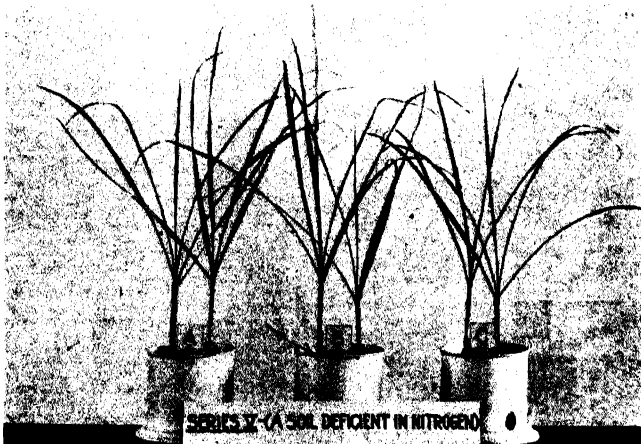


*A*—No weeds allowed to grow. *B*—Weeded at 6 weeks. *C*—Weeded at 8 weeks.





*A*—No weeds allowed to grow. *B*—Weeded at 6 weeks. *C*—Weeded at 8 weeks.





## Mineralizable Nitrogen in Some Hawaiian Soils\*

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With the increasing interest in the relation between utilization of nitrogen and growth and development of economic plants in Hawaii, attention has apparently been centered upon the maintenance of suitable levels of nitrogen in the plant with little regard for the natural nitrogen-supplying ability of the soil in which the plants are grown. In general, considerably over 90 per cent of the soil nitrogen exists in organic compounds of such a nature that this element cannot be utilized by green plants. However, certain microbiological activities within soils bring about the decomposition of these compounds with the liberation of mineral forms of nitrogen (i.e., nitrate and ammonia compounds) which are readily utilized by economic plants.

Attempts have often been made to use the nitrate and ammonia contents of soils as measures of the nitrogen fertility of soils, but with indifferent success. True, such a method measures the available nitrogen at the time the samples are taken, but gives little information as to possible future release of available nitrogen as a result of microbiological activities. Further, the data accruing from determinations of nitrate and ammonia may be seriously obscured because of the influences exerted by such factors as irrigation, rainfall, soil management practices prior to sampling, and duration and conditions of storage of soil specimens before the analyses are made. The purpose of this investigation was to adapt some procedure for measuring not only the existing available nitrogen in soils but also that which is released as a result of soil biological activities during the period a crop is being grown.

### DEFINITIONS

Since there appears in the literature considerable confusion regarding the definitions and terminology applied to the changes brought about in the soil nitrogen compounds by microbiological activities, the following definitions are offered to facilitate discussion:

*Mineralization:* The decomposition of the organic compounds of nitrogen into ammonium salts and nitrates.

*Ammonification:* The decomposition of the organic compounds of nitrogen into ammonium salts. (This is one of the steps taking place in mineralization.)

*Nitrification:* The oxidation of ammonium salts to nitrites to nitrates.

*Denitrification:* The reduction of nitrates to ammonium salts or to gaseous nitrogen.

*Mineralizable Nitrogen:* In this paper the term mineralizable nitrogen is used

\* Contribution of the Department of Chemistry and Soils. Published with the approval of the Director of the Hawaii Agricultural Experiment Station.

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to denote the amount of organic nitrogen which is mineralized when a soil is incubated under standard optimum conditions in the laboratory.

### HISTORICAL

During the first quarter of this century numerous workers studied the microbiological activities of soils as a means of measuring their fertility. Apparently the majority of these studies were concerned with the mineralization of nitrogenous materials added to soils rather than the mineralization of the naturally occurring nitrogenous compounds. However, studies on nitrate accumulation, such as reported by Russell (5), give some indication as to the rate of mineralization of the nitrogen within the soils. Waksman (6) studied the "nitrification of the soils' own nitrogen" in the laboratory by incubating soils wetted to optimum moisture conditions for 30 days. The increase in nitrates caused by this treatment was found to indicate the forms of nitrogen present in a particular soil and the speed with which they were transformed into nitrates and thus made available to plants. However, in this study there was no consideration of the changes occurring in the ammonia contents of the soils. Richardson (3), Orchard (2), and others working at the Rothamsted Experimental Station (4) have measured the increase in the nitrate and ammonia contents of soils resulting from incubation under ideal conditions in the laboratory. This mineralizable nitrogen content of soils was found to be related to the nitrogen responses in field experiments and to the nitrogen content of the non-leguminous herbage of pastures.

### METHODS

The laboratory determination of the amount of mineralizable nitrogen of a given soil consists essentially in wetting the soil to optimum moisture content, incubating it under standard conditions, and determining the amounts of ammonia and nitrate nitrogen\*.

Since no information was available, a test was undertaken to determine the optimum moisture content of a soil for mineralization. Samples of one soil, maintained at different moisture contents, were incubated under standard conditions and the mineralizable nitrogen determined.

Percentage of moisture	Mineralizable nitrogen mg/100g.
38	7.8
48	13.1
58	13.4
64*	13.3
78	12.8
88	5.8

\* Moisture equivalent.

The optimum moisture content for mineralization appears, as indicated, to be similar to the range suitable for the growth of green plants. Since the moisture equivalent of a soil is relatively easy to ascertain, it was chosen as the standard point to which all samples were moistened before mineralization determinations.

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\* The essential features of the technique for determining mineralizable nitrogen were not originated by the authors; they were obtained from the workers in the Chemistry Department of the Rothamsted Experimental Station.

*Preparation of Sample for Incubation:* In order for a sample of soil to be moistened to its moisture equivalent, it is necessary to determine both the moisture content and the moisture equivalent. The suction method suggested by Bouyoucos (1) has proved to be rapid and satisfactory for this purpose. The method consists in pouring a properly prepared soil sample into a 5-cm. Buchner funnel to a depth of 2.5 cm. The funnel is then placed in a beaker and water added until it almost reaches the level of the soil. After 24 hours the funnel is placed in a suction flask and suction continued for 15 minutes after the excess water has been drawn off. The moisture content of this soil is considered to be the moisture equivalent.

The weight of soil "y" necessary in order to have 50 grams of oven-dry soil may be calculated from the following:

$$y = 50(1 + p/100)$$

where "p" is the percentage of moisture in the sample on the dry basis. The amount of water "W" which must be added to "y" grams of soil in order to bring the soil to the moisture equivalent "M" may be calculated from

$$W = \frac{M - P}{2}$$

It has been recommended by other workers that, when the soils are incubated, the addition of certain mineral salts may facilitate microbiological activities. Consequently, the procedure adopted called for adding 10 c.c. of 0.25 per cent solution of di-potassium phosphate as part of the liquid used for wetting the soils to the moisture equivalent.

The calculated amount of soil "y" is weighed out and placed on a square of glazed paper, 10 c.c. of nutrient solution added, and the soil thoroughly mixed by rubbing with the fingers. Then the additional water required (W-10) is added by means of a burette or Mohr pipette, the soil is again rubbed, and transferred to a 500-c.c. Erlenmeyer flask. A test tube containing 15 c.c. of 30 per cent potassium hydroxide is placed in the flask in an upright position, and the flask is tightly stoppered. The flask is incubated for 3 weeks at 30° C. It is advisable, during the period of incubation, to change the atmosphere in the flask every 3 days by blowing air into the mouth of the container.

#### *Estimation of Ammonia and Nitrate Nitrogen:*

After the period of incubation the soil is removed from the flask and the nitrate and ammonia nitrogen contents are determined. Any reliable method for the estimation of these soil constituents may be used.

#### *Estimation of Carbon Dioxide Evolved:*

If desired, the carbon dioxide evolved by the soil microorganisms during the period of incubation may easily be determined by the method of Orchard (2). The contents of the potassium-hydroxide-containing test tube are washed into a titration flask and the solution is neutralized, using approximate 2N hydrochloric acid until the color of phenolphthalein just disappears. The bicarbonates are then titrated using standard N/10 hydrochloric acid and either methyl orange or brom phenol blue as an indicator. It is advisable to incubate a blank if this determination is to be made.

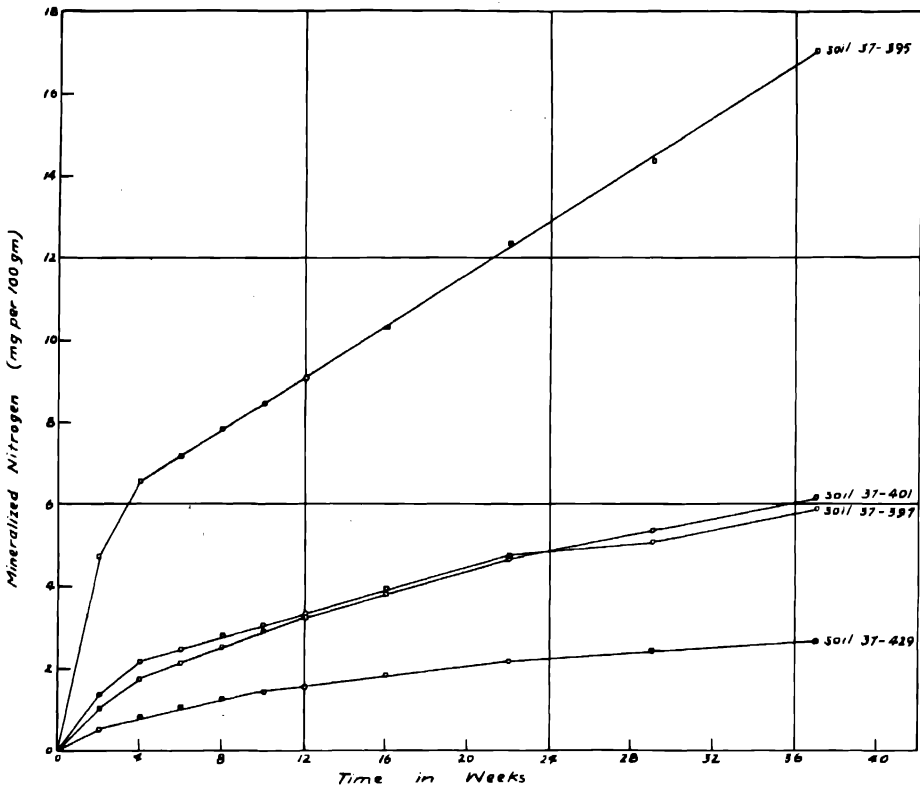


Fig. 1. The rate of mineralization of the nitrogen within soils.

#### THE RATE OF MINERALIZATION OF NITROGEN WITHIN THE SOIL

In Hawaii, many economic crops are grown on the same soil for long periods of time without disturbing the natural soil processes. Consequently, it is of interest to know something of the changes that take place in the rate of mineralization of the nitrogen within the soil when a sample is incubated over a long period of time.

Triplicate samples of four soils were incubated according to the previously described procedure. At intervals these soils were transferred into Buchner funnels and leached with 500 c.c. of water. The soils were then treated with 10 c.c. of nutrient solution and suction applied for 15 minutes after the solution had drained through them. The soils were replaced in their original flasks for further incubation. The combined ammonia and nitrate nitrogen in the leachates was determined. The results of these determinations for various intervals over a period of 37 weeks are presented in Fig. 1. These data suggest that the mineralization proceeds in two distinct phases; namely, an initial phase of rapid release of nitrogen taking place during the first 3 to 4 weeks of incubation, and a second phase having a much slower rate of release of nitrogen. The rate of release during the second phase of mineralization of three of the soils apparently decreases with time.

The ratios of carbon evolved as  $\text{CO}_2$  to nitrogen released as ammonium salts and nitrates during the various intervals of incubation were calculated, and are presented in Table I. These data show that during the first two intervals of incuba-

tion there was a much lower C/N ratio than during the remainder of the period. This suggests that during the initial rapid mineralization phase the microbiological activities of the soil are predominantly associated with nitrogenous compounds.

TABLE I

THE C/N RATIO OF THE C RELEASED AS CARBON DIOXIDE AND N RELEASED AS NITRATES AND AMMONIUM SALTS AT VARIOUS INTERVALS DURING A 37-WEEK PERIOD OF INCUBATION

Time in weeks	C/N ratio			
	Soil 37-395	Soil 37-397	Soil 37-401	Soil 37-429
0-2 .....	14.4	14.4	13.0	27.6
2-4 .....	15.8	11.2	16.4	35.8
4-6 .....	44.8	28.0	33.4	35.0
6-8 .....	37.0	22.4	32.2	34.8
8-10.....	35.8	21.6	30.6	29.2
10-12.....	37.0	24.2	41.8	50.0
12-16.....	41.0	26.6	37.2	40.8
16-22.....	32.4	21.0	34.0	32.0
22-29.....	39.6	28.8	46.8	55.6
29-37.....	36.2	24.6	36.4	52.8

#### MINERALIZABLE NITROGEN IN RELATION TO THE UPTAKE OF NITROGEN BY PANICUM GRASS

A series of soils was collected from various parts of the Territory for the purpose of comparing the total amount of nitrogen taken up by plants with mineralizable nitrogen contents of the soils. Duplicate 4-kilogram samples of each soil were potted, fertilized with 2 gm. of  $K_2HPO_4$  per pot, and 15 cuttings of panicum grass (*Panicum purpurascens*) planted. The grass, after growing in these pots for 78 days, was harvested, and the total nitrogen in the combined roots and tops determined. The scatter diagram, Fig. 2, shows the relationship between the mineralizable nitrogen in 4 kilograms of soil and the amount of nitrogen existing in the panicum grass which had been grown on the soil. A statistical analysis of the data showed a highly significant relationship to exist. It was found that the average utilization of the mineralizable nitrogen by the grass during the period of growth was 64 per cent.

#### SUMMARY

This paper deals with a study of mineralizable nitrogen in a number of Hawaiian soils, consideration being given to: (1) definitions, (2) determination of mineralizable nitrogen, (3) rate of mineralization, and (4) the relation between the mineralizable nitrogen and uptake of nitrogen by panicum grass; and may be summarized as follows:

(a) The moisture equivalent was shown to be a satisfactory moisture content at which to maintain soils when they are incubated for the purpose of determining mineralizable nitrogen.

(b) There appear to be two phases of nitrogen release when soils are incubated—an initial rapid phase (3 to 4 weeks), followed by a phase of slow release.



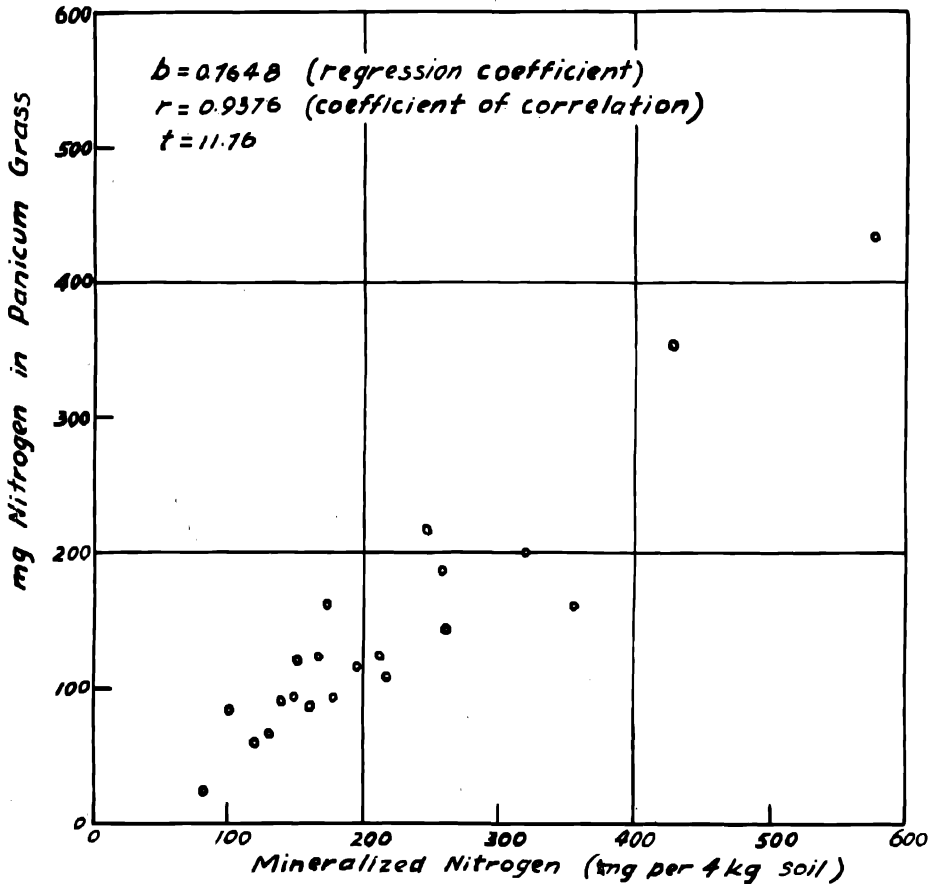


Fig. 2. Scatter diagram showing the relation between the uptake of nitrogen by panicum grass grown in pots to the mineralizable nitrogen of the soil.

(c) The initial phase appears to be associated with the microbiological decomposition of predominately nitrogenous compounds.

(d) A very significant relationship was found between the mineralizable nitrogen and the uptake of nitrogen by panicum grass grown in pots.

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## Plant Food Ratios for Sugar Cane Fertilizers

By R. J. BORDEN

Several years ago, when studying the tables of analytical data compiled by W. W. G. Moir\* from work previously reported by Stewart†, and Wolters‡, we were struck with the rather remarkable similarity of the ratio of  $N:P_2O_5:K_2O$  which was indicated by the chemical analyses of the H 109 sugar cane plants, not only when they had been harvested at different stages of their growth but also when they were taken from plots which had received different fertilizer treatments. For instance, when the percentage figures for nitrogen, phosphoric acid, and potash, which were found in the total dry weights from each individual harvest are set up in their approximate N:P:K ratio, we have evidence that such a ratio did not deviate very widely from 30:10:60 (3:1:6) in spite of the age of the cane or the plant food which it had received. Two tables which follow, summarize this evidence. (When the data in these two tables are compared, it must be remembered that the roots were included in the data of Table I but not in Table II):

TABLE I

(Data from analyses of crop from Oahu Sugar Company, Field 40, include total N,  $P_2O_5$ , and  $K_2O$  in total dry matter: tops, stalks, trash and roots.)

Approximate ratios of  $N:P_2O_5:K_2O$

Fertilizer treatment	At 1½ mos.	At 3½ mos.	At 5 mos.	At 8 mos.	At 12 mos.	At 17 mos.	At 24 mos.	Avg. for treatment
N	29:11:60	35:9:56	29:7:63	26:13:60	27:8:64	27:10:62	31:8:60	29:9:62
NP	27:13:59	33:8:58	29:7:63	27:14:58	30:10:59	28:10:61	28:8:63	29:10:60
NK	30:11:58	33:9:58	27:7:65	26:13:60	25:9:66	25:9:65	32:7:60	28:9:62
NPK	30:12:57	32:9:58	26:7:66	25:13:61	26:9:65	25:9:65	28:7:64	28:9:62
Average for age:	29:12:58	33:9:58	28:7:64	26:13:60	27:9:63	26:10:63	30:8:62	
Grand average—29:10:61								

TABLE II

(Data from analyses of crop from Pioneer Mill Company, Field 30, include percentages of N,  $P_2O_5$ , and  $K_2O$  from stalks, leaves, and tops only, i.e., no roots included.)

Approximate ratios of  $N:P_2O_5:K_2O$

Fertilizer treatment	At 6 mos.	At 12 mos.	At 15 mos.	At 21 mos.	Avg. for treatment
N .....	32:6:62	28:6:66	28:8:64	24:7:69	28:7:65
NP .....	27:9:64	24:9:67	27:9:64	21:9:70	25:9:66
NK .....	32:6:62	27:6:67	26:7:67	23:6:71	27:6:67
NPK .....	23:8:69	23:9:68	26:9:65	21:9:70	23:9:68
Average for age	29:7:64	26:7:67	27:8:65	22:8:70	
Grand average — 26:8:66					

\* Reports of Association of Hawaiian Sugar Technologists, pp. 189–192, 1930.

† Reports of Association of Hawaiian Sugar Technologists, pp. 199–220, 1929.

‡ Reports of Association of Hawaiian Sugar Technologists, pp. 141–159, 1929.

### *Detail of Present Study\*:*

Discussions, pro and con, of the probable value of an optimum ratio of the three principal plant foods for a complete fertilizer mixture have been common; specific evidence for such discussions has, however, been lacking. Difficulties are immediately apparent as soon as one undertakes to make a plan to secure this evidence, since with changes for the desired ratios, there will be changes in the total amounts of each nutrient supplied, and this factor itself may be largely responsible for the results which are secured.

In this attempt to secure comparable evidence, however, we planned a carefully controlled pot test, using three quite different soils which were potted, planted to POJ 2878 cane, and fertilized adequately with specific N:P:K ratios in triplicate. Since this was to be a study of N:P:K ratios, adequate amounts (particularly of nitrogen) for a normal crop were allowed for even the lowest figures of these ratios. The crop was harvested at the age of 12 months and the data which were secured have been summarized in the tables which follow.

#### *The Manoa Soil:*

The soil from Manoa was a residual, yellowish-brown silty loam, acid in its reaction (pH 5.6) and with a granular structure that makes it porous and well drained. It is known to be deficient in all three of the principal plant foods as well as in calcium. It has a very strong capacity to fix applied phosphates.

#### *The Makiki Soil:*

The alluvial soil from Makiki was a chocolate-brown loam, neutral in reaction (pH 7.2), with a much finer structure than the Manoa soil, and which drains more slowly when irrigated and tends to pack quite firmly, without cracking, however, as it dries. It is known to be well supplied with available phosphates and potash but is deficient in nitrogen.

#### *The Yamada Soil:*

The Yamada soil came from a field of brown-black clay loam alluvium which drains very slowly and cracks badly as it dries out. It has a neutral reaction (pH 6.9), an ample amount of both phosphate and potash but is deficient in its nitrogen supply.

#### *The N:P:K Ratios:*

Since it was physically impossible to include all of the suggested N:P:K ratios which we would have liked to test, we selected for certain groups which would enable us to study the effects of a change in the ratio of any two of these plant foods while the other nutrient was held at a constant. Our selection also afforded a chance to study the effect of a variation in one nutrient when the ratio of the other two was unchanged. We also included the 3:1:6 ( $1:\frac{1}{3}:2$ ) ratio which was suggested by the results shown in Tables I and II. Hence we have results which can be compared in the following groups:

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\* Experiment Station, H.S.P.A. Project A-105-No. 97.

Group I Nitrogen constant N:P <sub>2</sub> O <sub>5</sub> :K <sub>2</sub> O ratios		Group II Phosphate constant N:P <sub>2</sub> O <sub>5</sub> :K <sub>2</sub> O ratios		Group III Potash constant N:P <sub>2</sub> O <sub>5</sub> :K <sub>2</sub> O ratios	
1:0:4		1:1:3		1:3:1	
1:1:3		2:1:2		2:2:1	
1:2:2		3:1:1		3:1:1	
1:3:1					
1:4:0					
Group IV N and K constant N:P <sub>2</sub> O <sub>5</sub> :K <sub>2</sub> O ratio		Group IVa N and K constant N:P <sub>2</sub> O <sub>5</sub> :K <sub>2</sub> O ratio		Group V N and P constant N:P <sub>2</sub> O <sub>5</sub> :K <sub>2</sub> O ratio	
1:3:1		1:2:2		1:1:3	
1:½:1		1:¼:2		1:1:½	
				Group VI P and K constant N:P <sub>2</sub> O <sub>5</sub> :K <sub>2</sub> O ratio	
				3:1:1	
				½:1:1	

The pots were fertilized with their respective ratios of N:P:K from ammonium sulphate, superphosphate, and muriate of potash, once a month for the first eight consecutive months only. The total amounts used varied with the ratio to be provided, but unless a nutrient were to be omitted altogether, it was furnished for the lowest figure of a ratio, at an amount believed to be sufficient to grow normal cane; hence there was undoubtedly some luxury consumption when the higher amounts of a ratio were supplied.

#### Discussion:

The results are first presented and discussed separately for each of the three soils, and later the more pertinent comparisons from similar ratios on the different soils are summarized.

TABLE III  
THE MANOA SOIL  
(Figures are averages of three pots.)

Group	N:P:K ratio	lb cane	Y % C	lb sugar	Crusher juice analyses—				No. of stalks per pot	Wgt. per ft. of stalk (lb)	% of total stalks tasseled
					% Glucose	% N	% P <sub>2</sub> O <sub>5</sub>	% K <sub>2</sub> O			
I	1:0:4	3.3	11.0	.36	.52	.36	.007	.27	6	.26	0.6
	1:1:3	17.4	15.1	2.64	.45	.05	.03	.21	11	.41	16
	1:2:2	18.2	15.2	2.76	.48	.05	.06	.13	11	.39	12
	1:3:1	17.5	15.1	2.64	.66	.04	.09	.07	11	.39	0
	1:4:0	7.8	11.1	.87	.84	.13	.12	.03	7	.28	21
II	1:1:3	17.4	15.1	2.64	.45	.05	.03	.21	11	.41	16
	2:1:2	20.6	13.0	2.62	.57	.10	.02	.09	12	.41	13
	3:1:1	16.3	14.3*	2.33	.76	.19	.02	.07	13	.33	0
III	1:3:1	17.5	15.1	2.64	.66	.04	.09	.07	11	.39	0
	2:2:1	18.3	14.3	2.60	.63	.08	.05	.06	13	.36	0
	3:1:1	16.3	14.3	2.33	.76	.19	.02	.07	13	.33	0
IV	1:3:1	17.5	15.1	2.64	.66	.04	.09	.07	11	.39	0
	1:½:1	20.6	13.0	2.62	.57	.10	.02	.09	12	.41	13
IVa	1:2:2	18.2	15.2	2.76	.48	.05	.06	.13	11	.39	12
	1:¼:2	17.7	15.1	2.67	.50	.12	.007	.25	10	.42	7
V	1:1:3	17.4	15.1	2.64	.45	.05	.03	.21	11	.41	16
	1:1:½	18.3	14.3	2.60	.63	.08	.05	.06	13	.36	0
VI	3:1:1	16.3	14.3	2.33	.76	.19	.02	.07	13	.33	0
	½:1:1	18.2	15.2	2.76	.48	.05	.06	.13	11	.39	12

Amount of difference needed for significance:      2.1      1.3      .8

\* Only one juice sample, others lost.

*The Manoa Soil:*

When N is held at a constant (Group I) and the ratio of P to K is altered, we find no effect upon the cane yield or its quality, except when P or K is omitted entirely, in which case both yield and quality are adversely influenced. (It must be remembered that this soil is known to be deficient in phosphate and potash.) The percentage of P and K in the crusher juice was influenced directly and in accordance with the change in the P:K ratio.

With P held constant (Group II), the change of a 2 to 2 N:K ratio to either a 1 to 3 or to a 3 to 1 ratio caused a decrease in the cane yield which, however, had a better quality and hence gave no reliable differences in the sugar yield. Glucose and nitrogen in the crusher juice were increased with the higher nitrogen ratio, while potash also followed its change in the fertilizer ratio; phosphate was not affected. Cane weight per foot of stalk was somewhat less with the lowered potash ratio.

No reliable effect on yields or quality is noted from changing the N:P ratio when the K is held constant (Group III). Both nitrogen and phosphate in the crusher juice follow their changes in the fertilizer ratio, but glucose is not as definitely affected as it appeared to be with the higher nitrogen in the N:K ratios. There is an indication that the lower phosphate ratio has produced a somewhat lighter stalk weight.

With a 1:1 ratio of N and K (Group IV) a higher ratio of P to N and K (as 1:3:1) gave less cane, a better quality, and a similar sugar yield as a lower ratio (1:½:1). With a 1:2 ratio of N to K (Group IVa) there was no effect when a change in the ratio of P to N and K was made. Altering the N:P:K ratio from 1:1:3 to 1:1:½ (Group V) had no measurable effect. There was no significant amount of difference produced by changing the 3:1:1 ratio to ½:1:1 (Group VI). In these comparisons also, crusher juice analyses show positive relationships with the respective nutrient ratios that were supplied.

TABLE IV  
THE MAKIKI SOIL.

Group	N:P:K ratio	In cane	Y% C	In sugar	Crusher juice analyses—				No. of stalks per pot	Wgt. per ft. of stalk (lb.)	% of total stalks tasseled
					% Glucose	% N	% P <sub>2</sub> O <sub>5</sub>	% K <sub>2</sub> O			
I	1:0:4	16.1	15.4	2.48	.77	.04	.05	.25	16	.36	12
	1:1:3	16.8	14.5	2.42	.51	.05	.08	.23	10	.41	21
	1:2:2	15.6	14.5	2.23	.60	.03	.10	.23	12	.39	26
	1:3:1	13.4	14.8	1.99	.54	.03	.10	.18	10	.37	0
	1:4:0	12.9	15.2	1.93	.59	.04	.12	.05	12	.35	11
II	1:1:3	16.8	14.5	2.42	.51	.05	.08	.23	10	.41	21
	2:1:2	15.0	13.7	2.06	.53	.07	.08	.15	12	.34	0.4
	3:1:1	16.8	13.7	2.27	.89	.13	.09	.10	16	.36	0
III	1:3:1	13.4	14.8	1.99	.54	.03	.10	.18	10	.37	0
	2:2:1	13.0	13.8	1.79	.74	.10	.12	.11	11	.38	0.5
	3:1:1	16.8	13.7	2.27	.89	.13	.09	.10	16	.36	0
IV	1:3:1	13.4	14.8	1.99	.54	.03	.10	.18	10	.37	0
	1:½:1	15.0	13.7	2.06	.53	.07	.08	.15	12	.34	0.4
IVa	1:2:2	15.6	14.5	2.23	.60	.03	.10	.23	12	.39	26
	1:½:2	18.6	14.1	2.63	.54	.05	.06	.26	17	.40	12
V	1:1:3	16.8	14.5	2.42	.51	.05	.08	.23	10	.41	21
	1:1:½	13.0	13.8	1.79	.74	.10	.12	.11	11	.38	0.5
VI	3:1:1	16.8	13.7	2.27	.89	.13	.09	.10	16	.36	0
	½:1:1	15.6	14.5	2.23	.60	.03	.10	.23	12	.39	26

Amount of  
difference  
needed for  
significance:      4.7      .7      .64



### *The Yamada Soil:*

Contrary to expectations, the Yamada soil showed a response to both phosphate and potash; the reason for this is not apparent. When N was held constant (Group I) the low ratios of P to K produced slightly poorer cane yields; the cane quality was significantly poorer only when no phosphate was supplied.

With P constant (Group II) a change from a 2:2 ratio of N:K to either a 1:3 or a 3:1 ratio, gave poorer cane yields. With K constant (Group III) a 1:3 ratio of N to P shows increased cane yields over a 2:2 or a 3:1 ratio.

With a 1:1 ratio of N to K (Group IV) there was no reliable difference in cane or quality with changes in the P ratio; however, with a 1:2 ratio (Group IVa) of N to K the quality was better when P was high.

No differences in yield or quality are shown when the ratio of K to a constant N:P ratio is changed (Group V) but when the N ratio associated with a 1:1 P:K ratio (Group VI) is increased, both cane yield and quality are adversely affected.

Again, it will be noted that the percentage of N,  $P_2O_5$ , and  $K_2O$  in the crusher juice follow quite nicely their respective amounts supplied in the fertilizer. Glucose also followed the changes in the nitrogen ratios.

TABLE VI  
COMPARING THE PRINCIPAL YIELD VALUES OBTAINED FROM VARIATIONS IN  
THE N:P:K RATIOS ON THREE DIFFERENT SOILS

		Manoa soil		Makiki soil		Yamada soil	
		lb cane	Q.R.*	lb cane	Q.R.*	lb cane	Q.R.*
A:	(N:P:K) Ratios N:P+K						
	(1:3:1) 1:4	17.5	6.6	13.4	6.8	18.0	6.5
	(1:2:2) 1:4	18.2	6.6	15.6	6.9	17.9	6.3
	(1:1:3) 1:4	17.4	6.6	16.8	6.9	14.9	6.8
	(1:1/2:2) 1:2 1/2	17.7	6.6	18.6	7.1	16.8	7.5
	(2:1:2) 2:3	20.6	7.7	15.0	7.3	18.4	6.9
	(2:2:1) 2:3	18.3	7.0	13.0	7.3	14.6	6.9
	(3:1:1) 3:2	16.3	7.0	16.8	7.3	13.5	7.7
B:	(N:P:K) P:N+K						
	(1:1/2:2) 1/2:3	17.7	6.6	18.6	7.1	16.8	7.5
	(1:1:3) 1:4	17.4	6.6	16.8	6.9	14.9	6.8
	(2:1:2) 1:4	20.6	7.7	15.0	7.3	18.4	6.9
	(3:1:1) 1:4	16.3	7.0	16.8	7.3	13.5	7.7
	(1:2:2) 2:3	18.2	6.6	15.6	6.9	17.9	6.3
	(2:2:1) 2:3	18.3	7.0	13.0	7.3	14.6	6.9
	(1:3:1) 3:2	17.5	6.6	13.4	6.8	18.0	6.5
C:	(N:P:K) K:N+P						
	(1:3:1) 1:4	17.5	6.6	13.4	6.8	18.0	6.5
	(2:2:1) 1:4	18.3	7.0	13.0	7.3	14.6	6.9
	(3:1:1) 1:4	16.3	7.0	16.8	7.3	13.5	7.7
	(1:2:2) 2:3	18.2	6.6	15.6	6.9	17.9	6.3
	(2:1:2) 2:3	20.6	7.7	15.0	7.3	18.4	6.9
	(1:1/2:2) 2:1 1/2	17.7	6.6	18.6	7.1	16.8	7.5
	(1:1:3) 3:2	17.4	6.6	16.8	6.9	14.9	6.8

\* The reciprocal of the Y%C figures previously used in Tables III, IV, and V.

### *All Three Soils:*

If we attempt to bring together those N:P:K ratios which are associated with the higher sugar yield figures from the three soils, we have a picture like this:

On the Manoa soil	On the Makiki soil	On the Yamada soil
Ratio (lb sugar)	Ratio (lb sugar)	Ratio (lb sugar)
1:2:2 (2.76)	1:1/2:2 (2.63)	1:2:2 (2.85)
1:1/2:2 (2.67)	1:0:4 (2.48)	1:3:1 (2.77)
1:3:1 (2.64)	1:1:3 (2.42)	2:1:2 (2.63)
1:1:3 (2.64)		

Other than the fact that a low nitrogen ratio appears to be predominantly associated with the better sugar yields, it is doubtful whether any specific phosphate or potash ratios are similarly associated.

The evidence of these "best" ratios is certainly not in conformity with our knowledge of the sugar cane plant's requirements on these three soils. For instance, if the "best" ratio indicated for the Manoa soil is placed on a basis of pounds of each nutrient needed per acre, using 150 pounds of N as the most economical base, the complete fertilization would call for 150 pounds N, 300 pounds  $P_2O_5$ , and 300 pounds  $K_2O$ , and this would not be far from the known requirements. But on this same basis, the next best ratio would call for 150 pounds N, 50 pounds  $P_2O_5$  and 200 pounds  $K_2O$ , and this amount of  $P_2O_5$  and also of  $K_2O$  is considered inadequate for a satisfactory crop at Manoa.

On the Makiki soil, where at least 200 pounds of N is essential for a maximum yield, the "best" ratio would call for 200 pounds N, 66 pounds  $P_2O_5$  and 400 pounds of  $K_2O$ . This would provide an enormous waste of potash on this soil which is adequately supplied with potash from its natural sources.

Similarly, on the Yamada soil, where 200 pounds of N can be economically used, the "best" ratio would appear to call for 200 pounds N, 400 pounds  $P_2O_5$ , and 400 pounds  $K_2O$ ; again too much P and K.

Thus in practical parlance, sugar yields which were not significantly different were secured when ratios were used which would call for total applications (pounds of N,  $P_2O_5$ , and  $K_2O$  per crop) of approximately these magnitudes:

On the Manoa soil			On the Makiki soil			On the Yamada soil		
lb	lb	lb	lb	lb	lb	lb	lb	lb
N	$P_2O_5$	$K_2O$	N	$P_2O_5$	$K_2O$	N	$P_2O_5$	$K_2O$
150	300	300	200	66	400	200	400	400
150	50	300	200	0	800	200	600	200
150	450	150	200	200	600	400	200	400
150	150	450						

When a comparison of the relative effects upon cane yield and quality ratio secured from these three different soils is made in such a way that it shows the relationship between any single nutrient and the combined amounts of the other two nutrients (i.e., the ratio of N to P and K combined; of P to N and K combined; and of K to N and P combined) as in Table VI, there is little evidence to clarify an already complicated picture.

Even when we attempt to summarize the results of the major changes in a somewhat qualitative manner, as in Table VII, the data do not apparently furnish us with a reliable conclusion which we can evaluate and make practical use of, except perhaps that the results would appear to be the effects of a higher amount of N in the ratio.

From these tabulations, therefore, it is apparent that the effects are not always alike on different soils, and that it would be extremely hazardous to predict either an optimum ratio or the effect of any specific N:P:K ratio that might be supplied in a complete fertilizer mixture.



TABLE VII

SOME COMPARATIVE EFFECTS FROM THE MAJOR SPECIFIC CHANGES  
IN THE N:P:K RATIO

A—Condition: A change in the N:P:K ratio	Manoa soil		Makiki soil		Yamada soil	
	Cane yield	Quality	Cane yield	Quality	Cane yield	Quality
I.....from 1:3:1 to 1:1:3	=	=	+	—	—S	—
II.....from 1:1:3 to 3:1:1	—	—	=	—S	—	—S
III.....from 1:3:1 to 3:1:1	—	—	+	—S	—S	—S
IV.....from 1:3:1 to 1:½:1	+S	—S	=	—S	=	—
IVa.....from 1:2:2 to 1:½:2	=	=	+	—	—	—S
V.....from 1:1:3 to 1:1:½	+	—	—	—S	=	=
VI.....from 3:1:1 to ½:1:1	+	+	=	+S	+S	+S

B—Varied plant food ratios:				Effect on quality		
(Basic ratio 1:2:2)				On	On	On
Constant (1)	Increased (3)	Decreased (1)		Manoa soil	Makiki soil	Yamada soil
N	P <sub>2</sub> O <sub>5</sub>	K <sub>2</sub> O		=	+	=
N	K <sub>2</sub> O	P <sub>2</sub> O <sub>5</sub>		=	=	—
P <sub>2</sub> O <sub>5</sub>	N	K <sub>2</sub> O		+	=	—
P <sub>2</sub> O <sub>5</sub>	K <sub>2</sub> O	N		+S	+S	=
K <sub>2</sub> O	N	P <sub>2</sub> O <sub>5</sub>		=	=	—
K <sub>2</sub> O	P <sub>2</sub> O <sub>5</sub>	N		+	+S	+

**Legend:**

= No effect, i.e., less than the standard error.

— Poorer by an amount greater than the standard error.

+ Better by an amount greater than the standard error.

S Significant, i.e., greater than twice the standard error of a difference.

*Tasseling:*

The plants which were grown in this test afforded an opportunity to investigate the effect of various plant food ratios upon the tendency to produce tassels. The results are summarized in Table VIII. They indicate that a somewhat higher percentage of the stalks grown on the Yamada soil produced tassels than those grown on either of the other soils. The most apparent difference between these soils is one concerned with their physical characteristics—the Yamada soil being especially difficult to drain.

As a generalization, it would appear that there was a lower rate of tasseling when the ratio of K to N and P was low; with equal N and P, low K showed negligible tasseling.

With equal amounts of N and K, the results from high and from low P on the three soils were quite dissimilar. With a 1 to 2 ratio of N to K there was less tasseling with the lower amount of P.

With equal amounts of P and K, the higher amount of N showed a negligible amount of tasseling on all soils.

TABLE VIII  
PER CENT TASSELING

Group	N:P:K ratio	Manoa soil	Makiki soil	Yamada soil
I	{ 1:0:4	— 1	12	40
	{ 1:1:3	16	21	24
	{ 1:2:2	12	26	25
	{ 1:3:1	0	0	22
	{ 1:4:0	21	11	31
II	{ 1:1:3	16	21	24
	{ 2:1:2	13	— 1	12
	{ 3:1:1	0	0	— 1
III	{ 1:3:1	0	0	22
	{ 2:2:1	0	— 1	— 1
	{ 3:1:1	0	0	— 1
IV	{ 1:3:1	0	0	22
	{ 1:½:1	13	— 1	12
IVa	{ 1:2:2	12	26	25
	{ 1:⅓:2	7	12	8
V	{ 1:1:3	16	21	24
	{ 1:1:½	0	— 1	— 1
VI	{ 3:1:1	0	0	— 1
	{ ½:1:1	12	26	25
Average tassel . . . . .		8	9	18

*Summary:*

Studies of data previously collected had indicated a rather consistent and uniform ratio of N:P:K in the sugar cane plant.

The present study aimed to determine whether there might be such a thing as an optimum ratio of N:P:K which when used in a complete fertilizer mixture would give optimum yields.

Various ratios of N:P:K were tried on three quite different soils.

Results secured, in terms of sugar yields obtained, were not significantly different when some rather wide differences in the N:P:K ratios were used.

Those ratios which were found to be associated with the "best" yields were not generally in conformity with the known requirements of the three soils which were studied. Thus there is little encouragement brought forth that an optimum N:P:K ratio for a complete fertilizer mixture can be specified.

Crusher juice analyses showed a positive relationship with the respective nutrient ratios which were supplied. High glucose was associated with high nitrogen in the juice, which in turn was the effect of high nitrogen in the fertilizer ratio.

The effects of the various ratios upon tasseling indicated lower percentages of tasseling (1) when K was low in proportion to N and P, and (2) when N was high in relation to P and K; hence a suggestion that low potash and high nitrogen provide a nutrient relationship which is not conducive to abundant tasseling.



## Colloids in the Sugar Mill

By HUGO P. KORTSCHAK

In recent years it has been recognized that many of the difficulties of raw sugar manufacturing are caused by the presence of material in the colloidal state. Armed with this knowledge, engineers and chemists, and the salesmen of patent cure-alls, have devised countless methods for removing these colloids or rendering them harmless. Few of these methods have achieved general recognition, and none is suitable for all purposes and conditions.

That this is so should be no cause for surprise. Our knowledge of the complicated and ever-changing mixtures which we call cane juices is very vague. Seldom do we know just what it is that we wish to remove, or even why.

Where do these colloids come from? The sources may be divided into three classes. First, those which exist in the juice of the cane plant itself, and those which come from the cane stalk, we will call "inherent colloids." Second, there are the "extraneous colloids," coming from all the material entering the mill in addition to the cane stalk. Third, in the processes of manufacture, colloids may be formed. These we will call "process colloids."

### I. INHERENT COLLOIDS

#### *Pectin:*

This is one of the most important substances which exists in the juice in colloidal form. The basic unit of all pectic bodies is pectic acid, a ring structure of four galacturonic acid groups. Galacturonic acid is probably formed in the plant by the oxidation of the sugar galactose, from which it differs only by the presence of a carboxyl instead of the terminal alcoholic hydroxyl group.

Pectin itself is the form found in solution in the juice of the plant. It differs from pectic acid in that some of the carboxyl groups are esterified with methyl alcohol, which amounts to 10-12 per cent of the total. In addition, acetic acid, galactose, arabinose, xylose, araban, and cellulose may be combined with the ring. Pectins from various sources differ in composition as far as these additional groups are concerned. Since this is so, the exact constitution of sugar cane pectin will not be known until analyses have been made.

In the plant, most of the pectin is combined with the cell walls. In the sugar beet, this "protopectin" is in the form of a calcium—magnesium salt (11). Various means may be used to release the pectin from the fiber; for instance, heating with water, dilute acid, or salt solutions. This makes it certain that some pectin will be thus set free during the process of extraction in the sugar mill, and added to that already naturally present in the juice.

The cane plant contains relatively little pectin, 0.2 per cent being about the average, as nearly as can be judged from the scattered data available, and 1 per cent about the maximum to be expected. This may seem to be a small amount, but its importance may be shown by the effect on the viscosity, a solution containing one

part in a thousand of pectin having a viscosity comparable to a 10 per cent sugar solution.

Recent work in this laboratory has shown that pectin will combine with an equal quantity of citric or aconitic acids. Since these are the chief organic acids of cane juice, some or all of the pectin, depending on the concentration relationships, will be in this form.

What effect this will have on the properties of the pectin, outside of a slight lowering of the viscosity, is not known.

Evidence has also been found that sugar cane pectin carries a small amount of silica, although this may be only an adsorbed impurity.

In alkaline solution, pectin is almost immediately hydrolyzed to pectic acid. In acid solution, this also takes place, but much more slowly.

A well-known property of pectin is its power of increasing the solubility of sucrose. Upon what characteristics of the pectin this depends is not known, but it may vary enormously. In jellies, one part of pectin may "carry" from 100 to 500 parts of sugar. Some workers believe that this power increases with increased methylation of the hydroxyl groups of the pectin nucleus.

Pectin may be removed from solution by alcohol, or high concentrations of salts, which exert a dehydrating effect. Heavy metals, such as lead, precipitate it. Calcium does not precipitate pectin itself, but the calcium salt of pectic acid is very insoluble, so much so that it is used for quantitative analysis. Calcium pectate is, however, more soluble in sugar solutions.

Any vegetable material entering the mill will contain some pectin, thus not only the cane itself, but (to a very small extent) the leaves and, of course, weeds and trash will contribute.

In the process of extraction, pectin will have a slight effect, in that the increased viscosity will make it more difficult to remove the last traces of juice from the bagasse. The treatment with hot water will extract some pectin from the fiber, as mentioned above. According to Farnell (12) the amount so extracted will be small.

When the juice is limed to alkalinity, the pectin is hydrolyzed to pectic acid, the reaction going to completion almost immediately when the juice is heated. The pectic acid reacts with calcium ions to form calcium pectate. The electric charge on pectin is negative both in alkaline and acid solution (18) so that there is no question of an isoelectric point in the pH range used in the sugar mill.

What the physical properties of this calcium pectate will be is hard to predict. Calcium and sodium ions are antagonistic as far as pectin solutions are concerned (14, 18). This means that each will tend to neutralize the effect of the other, and potassium will probably behave similarly to sodium. Stuewer (19) finds that magnesium pectate has greater powers of adsorption than the sodium or potassium salt, but the ratios vary widely with changes in concentration.

Despite its negative charge, which is also that of the calcium phosphate floc (9), some of the calcium pectate is doubtless absorbed by this floc and carried down with it. The major portion, however, remains in the juice. Fortunately, the viscosity of pectate solutions, although still great, is considerably less than that of pectin solutions of equal concentration. As an example, at room temperature a 0.1 per cent pectin solution will have a viscosity 1.30 times that of water; when hydrolyzed to sodium pectate, the relative viscosity falls to 1.15. Pectates as well as pectin will

combine with citric or aconitic acid, the viscosity decrease being proportionately the same in both cases.

In evaporation, the presence of pectic substances will have little effect, as the concentrations are too small to affect the vapor pressure appreciably. They do cause a lowering of the surface tension, which will increase foaming, and the higher viscosity makes efficient heat transfer more difficult.

In crystallization, the presence of pectates is very unfavorable. Not only is the actual solubility of sucrose increased, but higher viscosity will slow down the velocity of crystallization, besides slowing the cooling, because of the higher specific heat and less efficient heat transfer. The shape and the purity of the sugar crystals will also be affected.

#### *Pentosans:*

The pentosans or gums are polymers of the pentose sugars. Araban may be hydrolyzed to arabinose, xylan to xylose, etc. In the sugar cane, araban seems to be the chief pentosan, with xylan also present. As mentioned before, pentosans are combined in the pectin molecule; this may be one of the chief sources of these compounds in the cane juice. The investigation by Cook (9), as well as that of Smith (16), shows that gums in the juice increase after clarification, being extracted from the small fiber particles which are present. Free pentosans will naturally increase when the pectin is hydrolyzed, during which reaction they are released from combination.

As far as the effects of pentosans on the mill processes are concerned these are similar to those of pectin, but in lesser degree. Whether pentosans also increase the solubility of sugar has not, apparently, been experimentally established, but it seems most probable. Since the pentosans are not removed by clarification, while some of the pectin is, they may be of greater final importance.

The conditions of growth, as well as the variety of cane from which the juice comes can influence the quantity of gums present greatly. Diseased or damaged cane is particularly apt to form exceptional quantities of pentosans.

#### *Protein:*

The proteins of the cane juice are largely albumins. These substances are present mainly in the growing point and leaves, so that when tops enter the mill with the cane itself there is a considerable increase in the protein content of the juice. The albumins are coagulated by heat, and thus removed during clarification. The only influence of any importance that they can have is to increase the difficulty of filtration, due to the gel-like character of the coagulum. Such proteins as are not coagulated by heat will remain as lyophilic colloids, having much the same physical effects as the pentosans. To a considerable extent, they will be decomposed to the constituent amino acids which go into true solution.

Any protein has an isoelectric point. Most of these fall in the pH range of 4 to 6. The "salt effect" due to the presence of electrolytes may change the pH of the isoelectric point, so that measurements made in water do not apply to cane juice; nevertheless it seems certain that the isoelectric points of most proteins will be within the pH range used in the mill.

The coagulation of proteins can be very easily reversed in some cases, usually by the addition of alkali (1). Here is a reaction which might well occur during clarification. To demonstrate the complex nature of protein coagulation, we may mention that albumin which has been extracted with ether is not coagulated by heat (2).

#### *Wax:*

Wax is one of the most important colloids in normal juices. Since wax is hydrophobic, some as yet unknown substance (probably the gums) must act as a protective colloid to cause its dispersion in the acid juice. This will be aided by the raised temperature of extraction, which is high enough to melt the wax, so that it forms a colloidal emulsion.

Cane wax consists mainly of myricyl alcohol,  $C_{30}H_{61}OH$ , esterified with caproic, stearic, and palmitic acids. According to Bardorf and Ball (5, 3; also Smith, 17), there are two waxes present. One is green, acetone soluble, melting at  $52^{\circ}$ ; the other, alcohol soluble, melting at  $82^{\circ}$ .

Wax is, along with the fat, the major cause of the turbidity of clarified juice. According to Smith (16) and Bardorf (4), as much as 1 per cent of the solids of clarified juice may consist of wax, or, more correctly, of wax and fat.

The wax appears to be very little influenced by any of the processes, appearing apparently in the original state, as the main organic colloid in refined sugar. Judging from the difficulty of saponifying other waxes, it is probably left unchanged by clarification. Once deposited on any surface, it clings tenaciously and is difficult to remove. Thus, besides adding to the difficulty of filtration, the wax will also be deposited on the surface of the sugar crystals as they are formed, and continue on to the refinery.

#### *Fats and Fatty Acids:*

A considerable search of the literature has failed to reveal any recognition of certain substances present in cane juice as fats. They have always been treated as part of the cane wax. Whether or not one or even both of the "waxes" found is really a fat is not known. Since they were largely saponifiable, this is not only possible but probable.

These fats consist of glycerides of palmitic and stearic acids. That they are easily saponified is a fact of some importance, as in the process of clarification the fatty acids are set free, forming soaps (16, 17). It is well known that calcium soaps have little or no detergent power, and this would make it seem as if the soaps formed would not be very harmful. It must not be forgotten, however, that there is a considerable concentration of potassium ions in the juice. When both calcium and alkali ions are present in a soap solution, the properties of the soap depend upon the ratio of the concentrations of these ions. If there is an excess of potassium ions, the properties of the compound will be more or less similar to those of a pure potassium soap (13, p. 41). We must not lose sight of the fact that there are other ions in the solution, too, any of which may be antagonistic to calcium or potassium, or to any other. Thus, prediction as to what extent the properties of the soap will be more nearly like those of potassium or calcium becomes impossible. Probably different forms will predominate in different samples of clarified juice.

The fact that most of the soap may be removed by filtration is difficult to understand. Two explanations may be advanced. Either the calcium salt form predominates, and the compounds are coagulated into relatively large particles, or else the soap is adsorbed on the surface of fine particles of fiber or other suspended matter. That the soap is so adsorbed can be shown by the decrease in fat content of the juice when it is screened to remove the fiber particles. This tends to make the second explanation more plausible. Here again it is possible that either explanation may hold, depending upon the conditions.

The difficulties which will be caused by the presence of soap in the juices need little elaboration, since its common use is due to the power of dispersing insoluble "dirt" in water. Any possible effect on the crystallization of the sugar is unknown, except that some of the soap will be adsorbed on the surface of the crystals.

#### *Polyphenols:*

Those impurities in the juice which are usually called "coloring matter" and "tannins" are chemically so similar that they may well be treated together. They consist of polyphenolic compounds, largely of the catechin type. They are usually present in such small amounts as to be unimportant. When they are present in larger quantities, as is the case with some varieties of cane, combination takes place with traces of iron in the juice to form dark-colored inks. No other influence of importance, besides that on the color of the product, can be ascribed to these substances.

The major portion of the polyphenolic compounds is removed by lime clarification. They are probably adsorbed on the calcium phosphate floc as calcium salts.

The source of these compounds is, in the case of the anthocyan coloring matter, the colored portion of the cane. The tannins are also present in the interior and in the juice. Injury to the cane plant may cause a large increase in tannin content.

#### *Starch:*

Most cane varieties contain little or no starch, but in a few this carbohydrate reaches considerable proportions. During extraction the starch passes into the juice. A typical lyophilic colloid, its properties resemble those of pectin, except that (1) the effect on the viscosity is much smaller, (2) there is no combination with citric or aconitic acid, and (3) it is not hydrolyzed during clarification. Although starch has been blamed for the poor clarifying properties of some juices, no case seems to have been proven. It would seem that starch concentrations high enough to be troublesome are most unlikely to occur. In one case at least, where difficulties were ascribed to starch, another compound, a glucose decomposition product, was found to be the real cause of the trouble.

## II. EXTRANEOUS COLLOIDS

#### *Humus:*

Of all the extraneous colloids, those which may cause the greatest difficulty are those derived from the soil. That this is so has been made evident when very large quantities of earth have been allowed to enter the mill. This is not the place for an extended treatment of the colloid chemistry of soils, so only a brief discussion will be given.



Humus is the name given to all the decomposed plant remnants which are present in the soil. Its constituents will include everything from still recognizable portions of leaves to bubbles of entrapped carbon dioxide gas, an end product of organic oxidations. An important group of constituents are the humic acids. These are often present as insoluble calcium salts which become colloiddally soluble when the calcium is removed. This may possibly occur during extraction. Other portions of the humus are already present as hydrated colloidal particles, sometimes in the form of a film, coating the inorganic portion of the soil (10).

From the humus, lyophilic colloidal material passes into the juice, where it will exert the usual effects of such colloids, i.e., increase in viscosity, more difficult filtration, etc. Where humic acids are present, these will be precipitated by lime. Other compounds may not be so easily eliminated. Of course, any compound occurring in the plant, except the very unstable ones, will be found in the humus.

### *Inorganic Colloids:*

Coming largely from soil, the inorganic colloid content of juices can vary greatly. The oxides and silicates of iron and aluminum make up the bulk of this material. These substances form, in the most part, hydrophobic colloidal suspensions which are very easily broken. In the presence of hydrophilic colloids, organic or inorganic, however, these inorganic particles are coated with an adsorbed layer of hydrophilic material, and thus they have, for all practical purposes, the properties of the protecting colloid. In clays, especially, this is already the case with the soil in its original state. Like a lyophilic colloidal solution, a protected lyophobic solution cannot be precipitated by minute concentrations of electrolytes, as the neutralization of the electric charge still leaves the protecting adsorbed water layer to prevent coagulation.

The same statements will apply to any inorganic colloids which may be formed during clarification. Positive evidence of such formation is lacking, but since it is very difficult to prevent colloidal solutions from forming even under laboratory conditions, there can be no doubt that colloidal calcium phosphate, to name but one, must be present in clarified juice.

The great importance of the colloids derived from the soil lies in the possibility of enormous quantities entering the mill under exceptional conditions. In such a case, all other colloidal constituents of the juice lose their importance, except as protective agents. It is fortunate that most of the soil particles will not be of colloidal size, as a rule, so that they will settle, though slowly, and not contribute too greatly to the viscosity.

Such inorganic colloidal matter as reaches the crystallizer will be quite as deleterious as the organic. Considerable quantities of sucrose are adsorbed even by hydrophobic minerals in the colloidal state (6).

In water, the isoelectric point of the oxides and silicates is near neutrality. For alumina, for instance, it is at pH 8.1. Thus, when other stabilizing factors are not present, such colloids will flocculate during clarification. It must not be forgotten that such flocculation under isoelectric conditions may take place although hydrophilic colloids are present. Even when the exact conditions are known, it is not always simple to predict whether an isoelectric solution will coagulate. In such a complex mixture as cane juice, it is impossible.

*Other Colloids:*

The various classes of material which have been mentioned do not, by any means, complete the possible colloidal substances which may enter cane juice.

When the cane is burned, products of polymerization, decomposition and oxidation are formed, whose nature will vary in each case to produce results which cannot be predicted with the amount of information available at present.

When weeds enter the mill in large quantities, they, too, will contribute to the juice. Except in the case of unusual plants, this will involve only an increase in the amounts of pectin, pentosan, starch, protein and polyphenols, but the possibility of completely new complicating factors can never be ruled out except by a study of the plant involved.

Fertilizers, insecticides, and weed sprays must also be remembered, especially in connection with their influence upon the properties of the soil.

## III. PROCESS COLLOIDS

Many of the substances already discussed might be placed under this heading, since they are not in the colloidal state until after the process of extraction. However, it seems more convenient to consider the "mill processes" as beginning with the mixed juice.

*Glucose Decomposition Products:*

"Nef has shown that glucose in the presence of sodium hydroxide reacts to yield an equilibrium mixture containing at least 93 different compounds, and concluded that (1) the initial fragments undergo molecular rearrangement to form more stable compounds, (2) they may react with each other, one being oxidized and the other reduced, (3) they may combine or polymerize, and (4) they may react with other substances, such as oxygen, to form acids, etc." (13, p. 633.)

A very small amount of glucose is always present in solution in a specially active form. The rate of formation of this form increases proportionately to the hydroxyl ion concentration; it is increased 100 times for a 22° rise in temperature (8, 15). Thus, the rate of formation will be increased about a billion (10<sup>9</sup>) times when a glucose solution at room temperature and pH 5 is brought to 100° and pH 8. This active glucose appears to undergo all the reactions of ordinary glucose, only at a greatly increased rate. (No reference to an active form of fructose appears to exist; from theoretical considerations there should be one, similar in properties to the active glucose.)

The multiplicity of the decomposition products of glucose makes it difficult to evaluate their effect upon the juices. Many of them are truly soluble; for instance, glycolic, formic, and oxalic acids. Other products, such as glyceric acid and saccharinic acid, especially in the form of their calcium salts, may have more nearly colloidal properties. The glucose decomposition products include many of dark color, and many which form highly viscous solutions.

*Sugar Salts:*

Sucrose itself may be considered a "border-line case" as far as the distinction between colloidal and true solutions is concerned. The calcium salts, not only of sucrose, but of glucose as well, have even more the properties of colloids.

Any saccharate will not, of course, crystallize out as sucrose. Outside of this, and the greater activity of the salts as compared with the sugars themselves, the effect of the salts will be similar to that of other hydrophilic colloids. Only small amounts of lime salts will be present under the usual conditions of clarification, for the sugars, viewed as acids, are very weak indeed, almost as weak as water. Note, however, that active glucose is a much stronger acid than is the normal form.

#### IV. THE INFLUENCE OF COLLOIDS ON MILL PROCESSES

The presence of colloids in the sugar mill exerts an unfavorable influence from the first process to the last. In extraction itself the effect is relatively minor, being due to the increased viscosity and stickiness of the juice, which tends to cause more of it to remain with the bagasse. Throughout the manufacturing processes the raised viscosity is a great handicap, slowing down operations and increasing power requirements.

##### *Clarification:*

The colloid chemistry of clarification is both important and complicated. Due to increased viscosity, colloids increase the possibility of uneven liming. As a result of this, some portions of the juice will be temporarily raised to too high a pH, while in others the main precipitation will take place at a lower pH and lime concentration than was intended. This will cause variation in the physical, if not in the chemical, form of the floc, and thus cause changes in its powers of adsorption and "sweeping."

Lime is added for two largely independent reasons. First, to neutralize the acidity of the juice; second, to precipitate phosphate. It has often been said that lime precipitates many other acidic substances, such as oxalate and citrate. It is improbable, however, that the concentration of any acid, except phosphoric, reaches a point where the true solubility of the calcium salt is exceeded. (The presence of diseased or damaged cane, or of soil high in humic acids, may cause exceptions to this.)

The calcium phosphate precipitate is largely in the form of an acid salt. The electric charge of the phosphate is negative (7). Such a precipitate will carry down with it mainly positively charged substances. (The colloids in the juice are, of course, negatively charged.) This does not exclude the possibility of other insoluble calcium salts, which are not present in sufficient concentration to be precipitated by themselves, being removed by coprecipitation.

One often encounters the statement that the phosphate floc, as it settles, sweeps the juice clear of colloids. The crudest definition of colloids states that they will pass through filter paper, which disposes of this possibility. Coarser suspended particles will be removed in this way, but long before particles of colloidal dimensions are reached this has lost its efficiency, as can easily be seen by the amount of material which filtration removes from clarified juice.

It has quite commonly been accepted as a fact that some of the colloids in cane juice are coagulated by slight alkalinity. Since increasing alkalinity increases the negative charge on the colloids, this seems unlikely, except for such where, as with the proteins, the charge is due to ionization of an amphoteric substance and the isoelectric point is in the neighborhood of neutrality. When such an electrolyte is

on the acid side of the isoelectric point and the pH is raised, the positive charge becomes smaller, as the basic ionization is suppressed. This assumes that there has been no adsorption of negative colloids by the positive ones, making the total charge of these negative in conformity with the bulk of the colloids present. The isoelectric point of the albumins is too low (around pH 4.5) and they will be negatively charged even before the juice is limed.

The change of the juice to alkaline reaction will precipitate the hydroxides of aluminum and iron from solution. These hydroxides precipitate as slimy gels of great adsorptive power. As amphoteric substances, they will, in the moment of formation, bear a nearly zero charge, which will rapidly be made negative by the adsorption of negative ions and colloids. This adsorption may or may not cause the hydroxides to stay in such finely divided form as to remain in suspension. A part will, of course, be attracted to the negatively charged phosphate floc, and thus removed.

The heating of the juice hastens the formation of larger particles of the precipitate. In addition, the temperature rise causes a great decrease in viscosity. These influences combine to cause an increase in the rate of settling, as, according to Stokes' Law, this rate is proportional to the square of the radius of the particles, and inversely proportional to the viscosity. The main influence of the hydrophilic colloids upon clarification lies in their action opposing these two tendencies. Adsorbed on the surface of small particles, they prevent their coalescence, due both to the surrounding water shell and to the repulsion caused by the electric charges. Due largely to this same water shell which increases the volume occupied by them, they raise the viscosity.

Another effect of the rise in temperature is the melting of the wax and fat, the liquid droplets being broken and dispersed still further by the violent agitation of the juice in its flow from the heaters to the settling tanks.

The albumins being unstable toward heat, they are dehydrated and coagulated, forming a gel-like precipitate which is carried down by the phosphate floc.

#### *Filtration:*

Difficulties in filtration are caused less by increased viscosity than by the presence of the adhesive gels which are formed in clarification. These adhere to the fiber particles which form the filter, clogging the pores, and making the passage of liquid impossible. The presence of large amounts of finely divided suspended matter of any kind has the same result. In addition, the gels which are present may hold relatively large amounts of water, thus causing loss of the sugar which is dissolved in it. Since a gel may contain as little as one part in a thousand of solid material, a small amount of impurity may immobilize a considerable volume of water. This water must not be confused with the "bound water" of hydrophilic colloids, which seldom exceeds 2—3 times the volume of the solid core.

#### *Evaporation:*

The colloids have practically no effect upon the vapor pressure of the juice. Raised viscosity causes uneven heating, and local superheating, with resultant "bumping." Reduced surface tension causes foaming.

Due to the increasing concentration of the solution, such substances with which

the juice is saturated, for example calcium phosphate, will precipitate. Since these materials are present in extreme dilution, the precipitates are likely to remain in colloidal form.

### *Crystallization:*

After evaporation, the colloidal characteristics have greatly changed, largely due to the formation of new material by the decomposition and oxidation of glucose and other substances. (Among these products may be oxalic acid, in the form of the very insoluble calcium salt, sometimes found as scale on the boiler walls.) The colloidal constituents of the massecuite do not appear to have been investigated except from the standpoint of total colloid content.

The presence of colloidal material will influence the shape and size of the sugar crystals, as well as their purity. The last is lowered by solid material, mostly the fatty acid soap, adsorbed on the crystal surface (17), as well as by increasing the amount of molasses adhering to the crystals. It may be well to point out that, since the crystals grow slowly, material adsorbed on the surface does not necessarily remain there, but may be in the interior of the final crystal.

The mechanism by which lyophilic colloids prevent the crystallization of a considerable part of the sugar present is still a matter of controversy. Important as this is to the sugar industry, investigations of this subject have been largely empirical attempts to remove either colloids or sugar from the molasses. The surprising constancy of the sucrose content of molasses from different factories leads one to the belief that the problem may not be as complicated as it would appear to be.

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# The Availability of Insoluble Phosphates to Sugar Cane

By A. S. AYRES

## INTRODUCTION

Phosphates are present in soils in many forms and combinations. In agricultural soils these occur in part naturally and in part from the interaction of added phosphate fertilizers with various constituents of the soil. Where it has been the practice to employ insoluble phosphate fertilizers, these may be present to some extent in much the same forms in which they were originally applied. All of these soil phosphates are very insoluble, as is attested by the extremely low concentrations of this nutrient in water extracts of soils and in drainage waters, even when these have originated in fields abundantly supplied with phosphorus.

In 1933, P. L. Gow and R. R. Ward, at this Experiment Station, conducted an experiment which was designed to show to what extent some of the simpler insoluble compounds of phosphorus presumably present in soils and certain insoluble commercial forms of phosphate were available to sugar cane. This was accomplished by growing cane plants in quartz sand cultures in which the only sources of phosphorus available to the plants were those under consideration. The insoluble forms of phosphate studied by these workers were raw rock, commercial reverted, and the phosphates of iron, aluminum and calcium. In order to compare the availability of these insoluble forms of phosphate with soluble forms, pots receiving sodium phosphate and calcium superphosphate were included in the test. The results of the experiment showed that all of the insoluble phosphates, except raw rock, were equally available to sugar cane. Moreover, they were fully as available as the soluble forms. The plants, which were fertilized with raw rock phosphate, were found to grow slower during the first months of the experiment than the other treated plants, thus indicating, apparently, a lower availability of phosphorus from this source. Subsequently, however, as though phosphorus were being made available at a greatly increased rate, the plants treated with raw rock phosphate began to grow much more rapidly—more rapidly, in fact, than the canes in any of the other treatments.

In 1935 the writer was assigned the task of carrying this research a step further by determining the degree to which certain forms of insoluble commercial phosphates employed in the study just cited are available to sugar cane when grown in soil instead of in sand cultures. It is with this experiment that the present paper deals.

## EXPERIMENTAL

Since raw rock and reverted phosphates are the only insoluble forms of this nutrient which have been used to any great extent on Island plantations, it was decided to limit the experiment to the consideration of these two forms. To serve as a criterion of the growth resulting from the use of these phosphates, a series of pots in which calcium superphosphate was the source of the element was included in the study. The ability of the soil to furnish the cane with phosphate from its own natural supply was to be determined by inclusion of a series of pots to which no phosphate would be added. Acting upon the observation of Gow and Ward that



the phosphorus from raw rock phosphate becomes available to sugar cane slowly at first, but later at a more rapid rate, a treatment was included in the test which consisted of a mixture of raw rock and reverted phosphates. It was thought that if the situation observed in sand cultures by these men similarly obtained in the soil, the reverted phosphate would meet the immediate needs of the young plants and the raw rock phosphate the later requirements. In addition to these treatments, three others were included in the experiment, namely: superphosphate plus lime, reverted phosphate plus lime, and lime alone. The thought back of lining the soil was to determine, in the case of superphosphate, if the presence of a good supply of calcium in the soil would, upon the addition of the soluble phosphate, cause the formation of a relatively insoluble calcium phosphate which would be more readily available to the cane plant than the products of fixation which would result were the additional lime not present in the soil. Where lime was used in conjunction with reverted phosphate it was hoped to determine if the partial neutralization by the lime of the acid soil to be employed in the experiment would effectively retard the solution and resultant fixation of the phosphate in forms less available to the cane plant.

The selection of a soil suitable for the study was of primary importance. The vital requirement was that the soil be sufficiently low in available phosphate to allow possible differences in the availabilities of the phosphates employed in the experiment to be reflected by corresponding differences in cane growth. For, manifestly, if the soil already contains sufficient phosphate to meet the needs of the crop, the addition of more phosphate, regardless of its availability, will not result in greater growth except, possibly, through some secondary effect upon the fertility of the soil. This is a point which is apt to be lost sight of in the planning of field experiments comparing raw rock with the various forms of soluble phosphates. Unless it can be shown that the soil under test is actually deficient in phosphate, as measured by crop response to fertilization with this nutrient, the mere failure to obtain more cane with soluble forms of phosphate than with raw rock does not prove that the two types are equally available.

Upon being advised that the soil of none of the cane lands on this island (Oahu) was sufficiently low in available phosphate to insure response to fertilization with this nutrient, the writer obtained soil samples from a dozen places at the Manoa Valley substation from which the necessary quantity of soil could be obtained. The suitability of the soils of the several locations was then determined by chemical analysis of the specimens. In the end it was necessary to resort to the use of a subsoil (Field 37, Manoa) in order to be sure of response even to soluble forms of phosphate. The analysis of the soil thus chosen is shown below:

#### ANALYSIS OF THE SOIL (MANOA) USED IN THE EXPERIMENT

Determination	R.C.M.*	1% citric acid soluble
$P_2O_5$ . . . . .	Low	0.0019%
CaO . . . . .	...	0.015%
pH . . . . .	4.6	.....
Phosphate fixation . . . . .	90-90-20	.....

\* *Soil and Plant Material Analyses by Rapid Chemical Methods.* The Hawaiian Planters' Record, 40: 189-299, 1936.

$P_2O_5$ : by extraction of soil with 0.5 N HCl.

pH: by modified LaMotte method.

Fixation: Figures refer to the percentages of applications of 1500, 7500 and 15,000 pounds  $P_2O_5$  per acre-foot of soil fixed from neutral solutions of  $(NH_4)_2HPO_4$ .

Available for this study was a set of 32 large (2' x 2' x 2') concrete pots which were set in the ground to a point a few inches from their rims. Drainage was provided by openings at the base of the pots through which drainage water might escape and be carried off.

After the soil selected for the experiment had been air dried and screened ( $\frac{1}{2}$ -inch mesh) the pots were filled to a depth of one foot and the soil thoroughly tamped down. All of the pots but those which were to receive lime were then filled to within 2 inches of the rim with more of the untreated soil. In the case of the lime treatments, the top foot of soil was thoroughly mixed with lime, in a mechanical mixer, at the rate of 9 tons per acre-foot of soil and placed in the proper pots.\* Water was then added in order to bring about the reaction of the introduced lime with the acid soil. The limed soils were thus maintained in a moist condition for a period of  $3\frac{1}{2}$  months prior to fertilization and planting. For the sake of uniformity of treatment, the unlimed soils were also kept moist.

Shortly after the period allowed for the reaction between the soil and lime had elapsed, the top 6 inches of soil in the pots was dried and, in all but the control pots, removed, intimately mixed with the appropriate quantity and form of phosphate and returned to the pots. Phosphates were applied in this manner at the rate of 500 pounds  $P_2O_5$  per acre-foot, based upon the surface area of soil in the pots. The eight treatments comprising the test were as follows (32 pots, allowing 4 replications of each treatment) :

- |                       |  |
|-----------------------|--|
| 1. No phosphate       | 5. Reverted and raw rock phosphates in equal amounts |
| 2. Superphosphate     | 6. Lime — no phosphate                               |
| 3. Reverted phosphate | 7. Lime — superphosphate                             |
| 4. Raw rock phosphate | 8. Lime — reverted phosphate.                        |

The pots were planted with H 109 seed on August 2, 1935. Three weeks later the stand was thinned to 4 plants per pot. Nitrogen and potash in the form of Chile potash nitrate were applied monthly, beginning August 31, 1935, or 4 weeks after planting. The initial application of this material was 50 pounds per acre of nitrogen and potash. Succeeding monthly applications were 100 pounds per acre of nitrogen and potash. The small amount of replanting that was necessary was done at the age of 5 weeks with pregerminated seed. The plants for this purpose were obtained from flats of black sand which had been planted at the time the experiment was begun.

#### DISCUSSION OF RESULTS

##### *Notes on Growth—Symptoms of Phosphate Deficiency:*

Response to phosphate fertilization was apparent within 2 months from the time of planting, the treated canes making a more rapid growth and exhibiting a

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\* Preliminary laboratory tests showed that this amount of agricultural lime was required to bring the pH of distilled water, mechanically agitated with the soil for 10 days, to the neutral point,  $pH = 7.0$ . Six months after the lime was mixed with the potted soil, during which time nitrogen and potash were applied in amounts totaling 250 pounds each of N and  $K_2O$  per acre from Chile potash nitrate, the reaction of the soil (in the limed controls) was  $pH = 6.0$ . At the same time that of the unlimed controls was  $pH = 4.8$ .



Fig. 1. Of the three pots in the foreground, the two containing four stalks each received no phosphate. The third pot received superphosphate. Note complete lack of secondary growth in the absence of added phosphate. Even at harvest only the four original stalks were present in the controls.

greener color than the controls. The lack of adequate phosphate in the controls was also evidenced by the complete failure of the canes in 7 of the 8 control pots to develop secondary growth. Development of secondary growth in the eighth control was confined to two of the four plants and was of a minor nature. This effect of phosphorus deficiency characterized these plants throughout the entire period of the experiment, only the 4 original stalks, with the exception noted, being present in each control at harvest. This point is illustrated in Fig. 1. At no time during

the experiment were any differences in growth apparent which could be attributed to the different forms of phosphate used. Lime appeared to be somewhat beneficial when employed together with phosphate.

*Procedure at Harvest:*

The cane from all treatments was harvested September 9-11, 1936, at the age of 13 months. Tops and millable cane were weighed separately and samples of each taken for analysis for phosphorus. Other portions of the stalks were ground and the resulting juice set aside for the determination of purity. The top 6 inches of soil, with which the phosphate had been mixed at the beginning of the experiment, was sampled for the determination of available phosphate.

*Availability of Soluble and Insoluble Phosphates as Measured by the Yield of Cane—Effect of Lime on Cane Yield:*

The weights of stalks and of tops harvested are shown in Table I. Referring to the table and considering first the unlimed soils, it will be seen that, judged by the growth produced, reverted and raw rock phosphates were about equally available to the cane plant. Moreover, these insoluble forms proved to be equally as available as (soluble) superphosphate. The response to all three forms of phosphate was very large, the yields in the treated soils being ten times those in the controls.

It will be noted that the findings of the present study are not in complete agreement with those of the experiment of Gow and Ward as regards the availability of raw rock phosphate. It is probable, however, that the highly acid soil employed in this test possessed greater power to bring this form of phosphate into solution than did the medium of the earlier experiment. Gow and Ward maintained the reaction of their nutrient solution between  $\text{pH} = 7.0 - 8.0$ . This suggests that while raw rock phosphate may be equally as available as soluble forms on acid soils, the same may be true on neutral or basic soils.

As in the unlimed soil, the insoluble phosphates proved to be equally as effective as the soluble form, so in the limed soils the single insoluble phosphate used (reverted phosphate) produced fully as much cane as superphosphate. The incorporation of lime into the soil did not, in the absence of added phosphate, result in significantly increased growth. However, where phosphate also was used, lime appears to have been beneficial. Thus, it will be observed in Table I that the average yields for super and reverted phosphates on the limed soil are considerably larger than for corresponding treatments on the unlimed soil. In the case of superphosphate, analysis of the data shows that the difference is probably a real one, while with reverted there seems no question but that increased growth resulted from the use of lime.

*Effect of Lime on the Quality of the Juice:*

In numerous experiments in the field large applications of lime to the soil have resulted in a marked lowering of the quality of the juice. In order to determine to what extent, if any, the gains in yield resulting from the use of lime in the present experiment were offset by poorer juices the purity of the juices from all treatments but the controls were determined.\* The results of the analyses are shown in

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\* Insufficient cane was present in the controls for the determination of both  $\text{P}_2\text{O}_5$  and juice quality. The former information was deemed more essential to the study.

TABLE I  
GREEN WEIGHTS OF TOPS AND STALKS (LBS.)

Treatments*	Pot	Tops	Stalks	Pot	Tops	Stalks	Pot	Tops	Stalks	Pot	Tops	Stalks	Avg.	Tops	Stalks
No phosphate .....	A-1	0.94	1.69	B-1	1.13	2.56	C-1	0.94	2.00	D-1	0.81	2.06	1	0.96	2.08
Superphosphate .....	A-2	7.56	22.38	B-2	7.38	19.56	C-2	7.31	24.38	D-2	4.00	21.31	2	6.56	21.91
Reverted phosphate .....	A-3	8.31	22.06	B-3	8.31	26.75	C-3	6.88	25.31	D-3	6.25	17.56	3	7.44	22.96
Raw rock phosphate .....	A-4	4.69	18.38	B-4	6.63	21.88	C-4	6.25	23.50	D-4	6.81	21.75	4	6.10	21.38
Reverted + raw rock phosphate† ..	A-5	8.31	20.56	B-5	6.31	17.63	C-5	6.94	21.19	D-5	5.81	18.38	5	6.84	19.44
Lime — no phosphate .....	A-6	1.00	1.75	B-6	1.31	2.69	C-6	1.44	2.94	D-6	1.06	2.13	6	1.20	2.38
Lime — superphosphate .....	A-7	8.75	24.38	B-7	7.25	29.94	C-7	8.00	25.13	D-7	6.88	22.88	7	7.72	25.58
Lime — reverted phosphate .....	A-8	9.69	35.31	B-8	8.00	32.81	C-8	8.81	25.00	D-8	6.63	33.50	8	8.28	31.66

\* 500 lbs. P<sub>2</sub>O<sub>5</sub> per acre based on surface area of soil in pot.  
† 250 lbs. P<sub>2</sub>O<sub>5</sub> each.

\* 500 lbs.  $P_2O_5$  per acre based on surface area of soil in pot.

† 250 lbs.  $P_2O_5$  each.

TABLE II  
APPARENT PURITIES OF THE JUICES AT HARVEST

Treatment	Pot	Purity	Pot	Purity	Pot	Purity	Pot	Purity	Avg.	Purity
No phosphate.....	A-1	*	B-1	*	C-1	*	D-1	*	1	*
Superphosphate.....	A-2	89.76	B-2	89.61	C-2	88.66	D-2	91.00	2	89.76
Reverted phosphate.....	A-3	88.56	B-3	90.06	C-3	89.62	D-3	89.15	3	89.35
Raw rock phosphate.....	A-4	90.66	B-4	92.17	C-4	88.44	D-4	88.03	4	89.83
Reverted + raw rock phosphate.....	A-5	88.57	B-5	90.57	C-5	88.37	D-5	87.96	5	88.88
Lime† — no phosphate.....	A-6	*	B-6	*	C-6	*	D-6	*	6	*
Lime — superphosphate.....	A-7	88.15	B-7	86.76	C-7	88.44	D-7	86.69	7	87.51
Lime — reverted phosphate.....	A-8	90.84	B-8	88.31	C-8	88.55	D-8	88.04	8	88.94

\* Insufficient juice for the determination of purity.

† Agricultural lime—at the rate of 9 tons per acre to pots 6-8, inclusive.

Table II. Since the cane was ground in a small experimental mill, these data are not comparable with the results obtained by methods of extraction which are more complete. However, they serve well enough to compare the effects of the different treatments. Reference to the table fails to reveal any clear-cut effect of treatment upon the quality of the juice. In this experiment the soils were only limed to about  $\text{pH} = 6.0$ . It is possible that the effect of lime on the juice is only detrimental beyond a point not reached in this study.

*Effect of Fertilization With Phosphate Upon the Percentage of Phosphate in the Plant:*

Reference to Table III will show that the percentage of phosphate in the dry matter of the tops was increased by the applications of this nutrient to the soil. This was true for all forms of phosphate used and on both the limed and the unlimed soils. The stalks of the treated plants, however, failed to show a corresponding effect of fertilization with phosphate. At first glance this result may seem to be at variance with many of the results of juice and stalk analyses of canes from phosphate experiments (2). In general, but by no means always, these have shown higher percentages of phosphate with increasing applications of the nutrient.

In the present instance, the failure of fertilization with phosphate to result in higher percentages of the nutrient in the stalks is probably explainable upon the basis of the extremely phosphate-deficient and high fixing soil used in this experiment, and the moderate applications of phosphate made to the soil. While the increases in available soil phosphate resulting from fertilization were sufficient to supply the canes with ten or more times the quantity of phosphate available to the control plants, thus making possible corresponding increases in growth, yet they were apparently insufficient to allow both for the increased cane growth and at the same time for a measurable accumulation of phosphate in the stalks above the percentage level of that in the controls. In the field, on the other hand, it is probably safe to say that the majority of phosphate experiments are on areas which are not deficient in this nutrient and upon which further applications of phosphate, since they would not result in increases in cane growth, would tend to raise the percentage of the element in the plant. Higher percentages of phosphorus in the plant might also be expected to obtain on phosphate-deficient soils if the quantity of the fertilizer added were considerably more than enough to produce maximum growth. The principle underlying the foregoing discussion is stated by Russell (3) in essentially the following words: If growth is being retarded due to an inadequate supply of some nutrient, the first increments of the nutrient added give considerable increments of crop; subsequent ones give less. So, the percentage of the nutrient in the crop may be unchanged by the first increments, or even fall, as the nutrient supply increases. With subsequent increases, however, higher percentages of the nutrient in the crop may be noted.

The fact that the tops reflected the supplies of phosphate available to the crop while the stalks did not suggests that the leaves, or possibly the non-millable green-leaf section of the stalk, might prove to be more sensitive indicators of the phosphate status of the soil than the millable stalks or juice.

TABLE III  
PERCENTAGE OF PHOSPHATE ( $P_2O_5$ ) IN DRY MATTER OF TOPS AND STALKS

Treatment	Pot	Tops	Stalks	Pot	Tops	Stalks	Pot	Tops	Stalks	Pot	Tops	Stalks	Avg.	Tops	Stalks
No phosphate.....	A-1	0.13	0.037	B-1	0.14	0.029	C-1	0.12	0.029	D-1	0.12	0.033	1	0.13	0.032
Superphosphate.....	A-2	0.16	0.029	B-2	0.15	0.028	C-2	0.17	0.029	D-2	0.18	0.039	2	0.17	0.031
Reverted phosphate.....	A-3	0.16	0.029	B-3	0.18	0.024	C-3	0.19	0.029	D-3	0.18	0.035	3	0.18	0.029
Raw rock phosphate.....	A-4	0.22	0.035	B-4	0.18	0.029	C-4	0.18	0.029	D-4	0.17	0.029	4	0.19	0.031
Reverted + raw rock phosphate.....	A-5	0.18	0.031	B-5	0.18	0.033	C-5	0.17	0.029	D-5	0.17	0.037	5	0.18	0.033
Lime — no phosphate.....	A-6	0.13	0.039	B-6	0.12	0.030	C-6	0.13	0.031	D-6	0.12	0.034	6	0.13	0.034
Lime — superphosphate.....	A-7	0.18	0.031	B-7	0.15	0.029	C-7	0.18	0.037	D-7	0.19	0.038	7	0.18	0.034
Lime — reverted phosphate.....	A-8	0.19	0.029	B-8	0.18	0.031	C-8	0.18	0.033	D-8	0.17	0.032	8	0.18	0.031

TABLE IV  
AVAILABLE (R.C.M.) PHOSPHATE IN THE SOILS AT HARVEST

Treatment	Pot	$P_2O_5$	Pot	$P_2O_5$	Pot	$P_2O_5$	Pot	$P_2O_5$	Pot	$P_2O_5$
No phosphate.....	A-1	Doubtful	B-1	Low	C-1	Low	D-1	Low	D-1	Low
Superphosphate.....	A-2	Medium	B-2	Doubtful	C-2	Doubtful+	D-2	Doubtful	D-2	Doubtful
Reverted phosphate.....	A-3	Medium	B-3	Doubtful	C-3	High+	D-3	High+	D-3	High+
Raw rock phosphate.....	A-4	High+	B-4	Medium+	C-4	High+	D-4	High+	D-4	High+
Reverted + raw rock phosphate.....	A-5	High+	B-5	High	C-5	High+	D-5	High+	D-5	High+
Lime — no phosphate.....	A-6	Low	B-6	Low	C-6	Low	D-6	Low	D-6	Low
Lime — superphosphate.....	A-7	Doubtful	B-7	Doubtful	C-7	Doubtful	D-7	Doubtful	D-7	Doubtful
Lime — reverted phosphate.....	A-8	Doubtful	B-8	Medium+	C-8	Doubtful+	D-8	Doubtful	D-8	Doubtful

*Discussion of the Results of the Soil Analyses at Harvest:*

The analyses of the soils of the various treatments at harvest indicated supplies of phosphate ranging from "low" to "high plus" (see Table IV). The values of "low" were confined entirely to the controls. A result of "doubtful" for Control A-1 can not be explained. Since, however, the other 7 controls gave values of "low" for available phosphate, we shall presume that this value represents the true status of this element in the untreated soils. It will be noted in the table that there is a tendency for the soils treated with reverted phosphate to give higher values than those which received superphosphate. Where raw rock phosphate was used, either alone or in combination with reverted phosphate, the results are unquestionably higher. Now, reference to Table V will show that the plant material harvested from the various phosphate treatments (on the unlimed soil) contained about equal quantities of phosphorus. It seems reasonable to assume, therefore, that the roots and accumulated dead leaves from the various treatments contained approximately equivalent amounts of this nutrient. If this is true, then quantities of phosphorus of the same order were removed from the several phosphate treatments on the unlimed soils. Now, identical quantities of phosphate fertilizer were applied to these soils. Therefore, the different values obtained for soil phosphate at harvest can mean one of two things:

First, that part of the superphosphate was leached out of the top six inches of soil, with which it was mixed, to a greater degree than was either reverted or raw rock phosphate, with the result that this layer of soil at harvest contained less phosphate in the case of the superphosphate treatment than where the insoluble forms were used. In the light of the writer's study of phosphate fixation in Hawaiian soils (1), the high fixing power of the soil used in this experiment and the intimate mechanical mixing of the soil and superphosphate, it seems unlikely that an appreciable amount of the superphosphate was leached below the zone in which it was originally placed. Moreover, even if we were to admit the possibility of leaching of superphosphate in the unlimed soil, we should certainly expect such action to be arrested by the presence of calcium in the limed soil. Yet, reference to the superphosphate treatments in Table IV fails to indicate more phosphate in the fertilized zone of the limed soil than in that of the unlimed soil.

The second possibility is that the added phosphates were present in the soil at harvest, in part at least, in different forms—in forms either not equally soluble, or not soluble at the same rate, in the R.C.M.  $P_2O_5$  reagent. To the writer this theory, that the different values obtained indicate not varying *amounts* of soil phosphate, but rather different *forms* of phosphate, seems the more plausible explanation.

It appears to follow, from this reasoning, that the higher R.C.M. values obtained on the soils treated with insoluble phosphates indicate the presence of greater or lesser portions of these fertilizers in much the same forms in which they were originally added to the soil. The lower values obtained on the superphosphate treated soils would then correspond to the solubility in the R.C.M. reagent of "fixed" phosphate, for there can be no doubt but that all but the minutest trace of the superphosphate would have been fixed under the conditions of the experiment months prior to the time at which the soils were sampled. If this postulate is correct, namely, that raw rock, and to a lesser extent, reverted phosphate were dissolved and fixed only in part after a year in a soil as acid as this one, and with



TABLE V  
GRAMS  $P_2O_5$  IN STALKS AND TOPS, PER POT

Treatment	Pot	$P_2O_5$	Pot	$P_2O_5$	Pot	$P_2O_5$	Pot	$P_2O_5$	Pot	$P_2O_5$	Avg.	$P_2O_5$
No phosphate	A-1	0.21	B-1	0.26	C-1	0.20	D-1	0.21	1	0.22		
Superphosphate	A-2	2.03	B-2	1.92	C-2	2.12	D-2	1.96	2	2.01		
Reverted phosphate	A-3	2.16	B-3	2.46	C-3	2.34	D-3	2.00	3	2.24		
Raw rock phosphate	A-4	1.88	B-4	2.05	C-4	2.13	D-4	2.08	4	2.03		
Reverted + raw rock phosphate	A-5	2.35	B-5	2.01	C-5	2.07	D-5	2.01	5	2.11		
Lime — no phosphate	A-6	0.30	B-6	0.28	C-6	0.30	D-6	0.25	6	0.28		
Lime — superphosphate	A-7	2.66	B-7	2.29	C-7	2.73	D-7	2.64	7	2.58		
Lime — reverted phosphate	A-8	3.35	B-8	3.04	C-8	2.61	D-8	2.78	8	2.95		

which the phosphate was intimately mixed, then we should expect under the conditions of ordinary field practice that these phosphates would remain undissolved and unfixed for a much longer period of time.

On the basis of this interpretation of the results of the soil analyses, there appears to be no support for the theory, presented earlier in this paper, that the presence of the added lime might result in the wholesale precipitation by calcium of the added superphosphate. For, if such had been the case, the values for phosphate in the soils which received both lime and superphosphate would, if we may neglect the slightly greater absorption of phosphorus from this treatment, have been higher than on those which received superphosphate alone. Reference to Table IV shows that such was not the case. Neither do the data offer evidence that the lime retarded the solution and subsequent fixation of the reverted phosphate, as was thought might prove to be the case. In fact, there was some tendency for soil phosphate to be lower where lime was used in conjunction with reverted phosphate than where the phosphate alone was used. This may have been due to the greater absorption of phosphorus from the limed soil.

#### SUMMARY AND CONCLUSIONS

The availability to sugar cane of reverted and raw rock phosphates was compared with that of calcium superphosphate. These insoluble phosphates were found to be equally as available to sugar cane as superphosphate in the acid, high phosphate-fixing soil used in the experiment. All three forms of phosphate resulted in yields ten times those obtained in the controls. Even when the acidity of the soil was reduced by liming, insoluble reverted phosphate proved to be equally as available as superphosphate.

The effect upon the yield of liming the soil prior to fertilization with phosphate was determined both in the case of a soluble and an insoluble phosphate. Super and reverted phosphates on the limed soil resulted in larger yields than when these phosphates were employed without lime. Lime, in the absence of added phosphate, did not result in appreciably increased cane growth.

The addition of lime to the soil at the rate of 9 tons per acre was not found to adversely affect the quality of the juice.

Attention was given to the possible use of the percentage of phosphorus in the cane plant as an indication of the supply of this nutrient in the soil. Marked differences in the supplies of available phosphate in the soil did not result in differences in the percentages of this nutrient in millable cane. The tops, however, were found to reflect the supply of phosphate in the soil.

The symptoms of phosphate deficiency in the control canes were: (a) lack of normal growth, (b) yellowish color of the leaves, and (c) complete absence of tillering.

Markedly higher values for R.C.M. phosphate at harvest were found in the soils which had been fertilized with raw rock phosphate than in those which had received superphosphate. This is interpreted as indicating that a considerable portion, at least, of this insoluble phosphate was still present in the soil at harvest in much its original form. It is suggested that under field conditions the period required for the solution and subsequent fixation of raw rock phosphate would be much greater. To a lesser extent reverted phosphate was found to resist fixation.

The writer is indebted to the Sugar Technology department for the analysis of the juices. He also wishes to express his appreciation to R. J. Borden, P. L. Gow and D. S. Judd for counsel received and to Wm. SaNing for assistance in conducting the experiment.

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The Sixth Congress  
of the  
**International Society of Sugar Cane Technologists**  
October 24 to November 5, 1938  
at  
Louisiana State University, Baton Rouge, Louisiana

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By W. W. G. MOIR

*December 9, 1938.*

Experiment Station, H.S.P.A.,  
Honolulu, Hawaii.

*Dear sirs:*

Since I was sent to the Sixth Congress of the International Society of Sugar Cane Technologists in New Orleans and Baton Rouge, Louisiana, as a representative and chairman of the delegation, I feel it only right that some report or record be submitted of things seen or information secured. The report requested by the program committee of the Hawaiian Sugar Technologists is included herewith, as it contains more details and will save repetition. A copy of the reports of committees and abstracts of papers submitted, which were given each delegate upon registration, is also appended, as well as a pamphlet from the U. S. Industrial Alcohol Company.

The delegation from Hawaii consisted of Messrs. H. P. Agee, W. van H. Duker, C. R. Ferdun, L. D. Larsen, Toru Oishi, S. S. Peck, C. E. Pemberton, O. H. Swezey, Keith Tester, J. N. P. Webster, C. A. Wisner and the writer. Somewhere between 275 and 300 delegates attended the meetings, which probably made it the largest thus far held by the Society. The writer has attended five of the six congresses held by this Society, having missed the Australian gathering. Of those meetings attended, this one was probably the most outstanding insofar as the presentation of papers based on scientific research is concerned. Usually there have been a great many papers on general items of cane cultivation, but at this meeting there seemed to be a dominance of reports on scientific investigations. These were not confined to one or two countries but were presented from every country represented at the gathering. One of the outstanding differences of this congress from those of the past was the preponderance of younger men who enthusiastically, ably and unhesitatingly presented their work to the world for criticism or commendation.

Hawaii has for many years been the envy of other sugar producing countries in the great use it has made of research. Other countries have followed our policies and developed them to suit their particular needs. Instead of finding ourselves still out in front in the race for information on better cane culture and sugar production, we now must admit we are running a race where the other participants are on our heels, and running neck and neck in some matters. These, in general, are the conclusions one must arrive at after listening to the delegates from other countries, presenting the results of their investigations.

Eleven years ago, I visited Louisiana and saw a sugar industry that was about to close its doors but which had one ray of hope presented to it in the form of new canes. Today one finds a thriving industry with potentialities of production far exceeding quotas allotted by the A. A. A. In fact, under the sugar control program the industry doubled its output in the five years from 1933 to 1937. Even under the severe control of acreage, as well as quota, decreed for 1939, the crop may again exceed the quota and a large carry-over result. This phenomenal improvement in the state of an industry is almost entirely due to research.

As the result of the foreign introductions and cane breeding work carried on by Dr. E. W. Brandes and his associates in the Sugar Plants Investigation Bureau of the United States Department of Agriculture, together with the cooperation of the Experiment Station at Louisiana State University, mosaic disease has practically been conquered. With the control of this disease, a great stimulation has taken place in all lines of investigation on soils, fertilizers, harvesting and windrowing losses. A new outlook on crop production and hopes for the future have so heartened the planter that today one finds a naturally expanding industry creating rather serious problems for the sugar control program of the A.A.A.

The mainland cane growers will probably be forced to reduce their acreages in cane in 1939. The 1938 crop will fill their quota and produce a reserve from an acreage of about 325,000 acres. It has been estimated by the A.A.A. that 250,000 acres will produce the necessary quota for 1939. If these figures are correct, it will mean a 75,000-acre reduction, or about 24 per cent. This also means that there will be no second ratoons, and that, in turn, means there will be more cover-cropping. This will result in saving fertilizer costs on the plant crop, as well as producing a heavier crop. You will readily see that the chances for a much larger production are very strong and that the A.A.A. control is really necessary to prevent over-production.

Outside of the wonderful return of this industry to a place in the sun through the use of disease-resistant canes, much has been accomplished in cultural and other practices. The severe curtailment of germination in seed and stubble, after the winter dormancy, has been a marked control of crop production. The newer canes have aided in the remedy of this, but the new practice of planting in August instead of October and November will do even more. The better established root and stubble formation from earlier planting aids in withstanding the severe conditions of the winter weather.

With better varieties, fertilizer responses are now secured. These are mainly in ratoon crops. With these heavier-producing canes, harvesting operations have been greatly improved and costs reduced. The excellent use of cheaper and lighter equipment in harvesting operations greatly interested those from Hawaii and serious consideration should be given towards adapting these methods to Hawaii.

While visiting in Colorado, I had the opportunity to become acquainted with some of the field problems of the sugar beet industry. It was most heartening to find that the same basic principles of sugar production found in cane were applicable to beets. Sugar is laid down in greater concentration in the rings of the beet in a sort of hollow, inverted, conical area midway between the center of the beet and the outside layers. The fibro-vascular bundles of the older leaves are connected with the center core while those of the newest leaves are connected with the outer rings. The

rings laid down when sunshine, moisture and fertility are at their optimum are those producing the most sugar. The work of the late Dr. Das has shown that in sugar cane this is true of those joints laid down under optimum conditions. The quality and quantity of sugar produced is materially affected by the vigor of growth, the excess or deficiency of moisture, or the unbalancing of optimum conditions through disease, climate, or nutrition. This year, with heavy, late rains and heavy disease attacks, late growth was secured. Poor sugar storage resulted because of the poor weather and leaf spot disease. The yields of beets were good but the sugar content was low. Many points of similarity exist in the two sugar crops and this makes a visit to the beet area exceedingly interesting. The Grand Lake Water and Power Project now being started will bring irrigation water to these sugar beet growing areas. This will mean a normal crop each year instead of once in two or three. It also means the possible expansion and overproduction in this area.

Pen-feeding both cattle and sheep are very thriving operations on farms adjoining sugar beet factories. Beet pulp, molasses and soybean or cottonseed meal furnish the balanced rations. Cattle and sheep are purchased just before the beet harvesting starts and are sold at the end of the harvesting season. The cooperation, dependency, and mutual benefits secured by these cattle raisers and beet producers should be an incentive to sugar cane growers and ranchers in Hawaii to inaugurate similar practices.

In a discussion with J. B. Trinler, Manager of Central Preston, Cuba, it was found that his company has been having excellent results with pen-feeding cattle with a mixture of yeast, molasses and very fine powdered bagasse, all of which were produced at home.

The great interest shown by delegates at the meeting in the utilization of by-products of the sugar industry—the visit to the International plant of the U. S. Industrial Alcohol Company, and the Celotex plant—and the discussions held with experts on these matters, greatly impressed several of the Hawaiian delegates. More than once our members expressed their hopes that our local industry would embark on the development of more by-products on a large scale and that the Experiment Station be permitted to expand on the present investigations. This work would materially help our industry through these depressing times.

The excellent papers presented by delegates from all over the world gave detailed results of research that has materially aided crop production through disease and pest control, proper nutrition, and cheaper harvesting and milling operations. It is sincerely hoped that these papers will be reviewed in our Station publications and useful data analyzed for our education. It was exceedingly stimulating to see how the rest of the sugar producing countries had so strongly turned to research in the solving of their problems. This also strongly impressed on our minds the even greater need of research at our Station to maintain our existence in the ever keener competition. The heavier and cheaper production in other sugar growing countries, but more especially in those of our mainland domestic areas, makes even greater research imperative if we wish to survive in this economic war. Here's hoping we may ever keep our heads up and lead the way.

Respectfully submitted,

(W. W. G. M.)

This cane sugar congress, which was probably the largest thus far held, was most ably and efficiently organized. Great credit is due the hosts and hostesses for their hospitality and endless efforts to make this the very best congress. The writer feels that they were very successful in these efforts. An excellent change was made in the usual congress procedure so that after a preliminary get-together the delegates took a tour through the sugar section of Louisiana before getting down to the more serious work of listening to and discussing matters presented to the various sections of the society. On this tour each day's visits to plantations and communities showed us again and again the genuine Southern hospitality in all its perfection. During the first two days we visited the International plant of the U. S. Industrial Alcohol Company, the Gramercy Refinery of the Colonial Sugars Company, the Chalmette Refinery of the American Sugar Refining Company, and the Celotex plant of the Celotex Corporation before leaving New Orleans on our Greyhound tour of the sugar-growing parishes, but more especially of the Evangeline country and its bayous.

On one of the days, while still at New Orleans, a visit was made to Reserve Plantation owned by Godchaux Sugars. Several sugar factories were visited by the mill group on the tour, while the field group was taken to field tests on varieties, fertilizer and diseases, and to see general field operations.

To more thoroughly understand the methods and problems of the Louisiana sugar industry, a general but brief survey of history, soils, climate, labor and transportation should first be given.

From the introduction of sugar cane to Louisiana in 1751 down to the present time, one finds a most interesting story of the trials and tribulations of the early French and Spanish pioneers and the many others that have followed them. A chart of the sugar production per year will show you just how the ups and downs of their industry have followed the governmental affairs and disasters that normally befall most any agricultural crop. Probably the greatest and most rapid decline was that in the period from 1922 to 1926 and this was almost entirely due to the ravages of the mosaic disease. Likewise, the most spectacular increase was in the years from 1930 up to the present when resistant varieties, imported or created with the cooperation of the United States Department of Agriculture, have re-established a necessary industry. It can be said without fear of contradiction that without the use of disease-resistant or tolerant varieties the sugar industry of Louisiana would now be a thing of the past.

Louisiana is at best only a sub-tropical country and it is only due to the phenomenal growth secured from these newer varieties in the few good growing months of the year that a crop can be secured. Planting is carried out usually in the fall months of September and October. Ratooning follows the harvesting in November and December. Since wet and frosty weather usually exists up to the end of April, there is really but six to eight months of growing time left to produce a crop. During the months of July, August, and September cane growth is phenomenal. The two great drawbacks to heavy crops are, first the slowness and, second, the lack of proper stooling after the winter dormancy. In ratoons, and more especially in plant cane, this poor germination is a very serious curtailment. A new practice that has become more widespread and very promising is that of planting in August whenever body seed can be secured. This early plant will often be killed down

with the frost in winter but the stubble has become sturdier and better established than that from fall plantings. Long stalks are more often used for seed in contrast to short seed pieces. Depth of planting depends largely upon the time of the year when planting is done—that is, deeper for protection in cold weather.

Fertilization is varied according to soil type, variety, plant or ratoon and to individual likes and dislikes. It has become almost a uniform practice to fertilize ratoons, but not all plant crops are so treated. All fertilization is carried out in the late spring or early summer in one application when some stalk formation is showing. A single application has been found to be as effective as several. The practice of rotating cane with a cover crop, mainly soybeans or cowpeas, between the last ratoon crop and the next plant is almost universal. To those of us from Hawaii, this loss of one year in growing time for the opportunity of growing a plant crop without fertilization seems uneconomical. But with government control of production and much available land, the practice seems to have its merits for Louisiana. Since no second ratoons (A.A.A. decree) may be raised this next year, more land must be fallowed and cover cropping should materially reduce fertilization costs and stimulate heavier production.

In the spring, the first efforts must be directed towards aiding the germination of the eyes on the seed or stubble. This is usually done by the practice of off-barring to allow the heat of the sun to warm up the soil closest to the eyes. After germination has been secured, cultivation is resorted to to maintain proper tilth and control weeds. Many tractor-drawn implements are in use for these processes. The seed is originally planted in a single line on the top of six-foot beds between deep furrows. This is necessary to maintain proper drainage. As cultivation continues the cane becomes more hilled up and, from a steep-sided furrow and a rounded top to the bed at planting time, a change is made to a more rounded bottom to the furrow and a hilled-up row of cane on the top of the bed at the final cultivation. The furrows usually run in the direction of the greatest fall in slope. Since most of the land is so nearly flat it becomes quite a serious problem to secure proper drainage in certain areas. The rows are usually six feet apart and every hundred feet a deeper furrow or drain is installed for carrying away excess water. Cross drains, opening into these, cut the furrows at right angles. These are not much deeper than the furrows and occur at varying distances, depending upon the slope of the land and the extent of the planting. The cane rows are therefore found to be from 12 to 15 inches above the bottom of the furrow at harvest time. This permits faster harvesting operations; first, by making it easier to cut either by hand or by machinery, and second, furnishing a handy location to separate the trash from the cane. The cleaned cane stalks are laid from stubble to stubble on the two crests of adjoining cane rows over a furrow in which there is no trash. This greatly aids the loading of cane without interference from trash.

Deep plowing, that is deeper than 15 inches, is not often found but experiments under way at the plots of the Experiment Station show much better drainage and healthier cane where deeper plowing was tried. Some cover cropping is carried out by planting legumes on either side of the cane row and burying the crop in the spring before the cane closes in. This saves on some weeding. Often the only preparation a cane field gets before planting is the splitting of the old bed in two and the burying of the cover crop in the furrow on either side. After the cover crop has rotted, in



about one month's time, the planting proceeds on the new bed created where the old furrow existed.

Many points indicate that there might be grounds for their claims of the necessity of returning organic matter to these soils. These are (1) light soils, easily dried out, predominate, (2) very little trash decomposition, (3) only one plant and one ratoon crop and, therefore, not much root formation and its resulting decomposition, (4) all trash burnt off after harvesting, and (5) cold, wet weather part of the year so that organic matter cannot be turned into a useful material during part of the year and probably too rapid production at other times.

It must be remembered that yields of from 15 to 35 tons are the crops that are produced on these lands in the short growing season.

Much of the crop must be windrowed before the grinding season is over because of the fear of killing frosts. Not all varieties keep well in the windrow and not all varieties mature early enough for early harvesting. At the present time, C.P. 28-19 is the favorite for early harvesting but is rapidly being displaced by C.P. 29-320. Co. 281 is still the favorite cane for windrowing as it keeps remarkably well. Mosaic, however, is taking a terrific toll in this variety. C.P. 29-116 is a late maturer but does not windrow as well as Co. 281. Co. 290 is quite a favorite in black soils and C.P. 28-11 in low, wet lands. Both canes, however, are easily flattened with wind. C.P. 29-320 is very resistant to mosaic and will readily recover if infected material is planted. However, it is susceptible to chlorotic streak. C.P. 28-11 and 29-116 are resistant to root-rot troubles, while 29-320 is intermediate and Co. 281 and C.P. 28-19 susceptible and only succeed in light, well-drained soils.

To get a general idea of the soils of the sugar belt, a few excerpts from the abstract of the paper by O'Neal and Hurst on "The Soils of the Sugarcane District of Louisiana" are quoted herewith:

The soils of the sugar cane district of Louisiana are largely of alluvial origin and have been derived from rich sediments brought down by the Mississippi River and its tributaries during periods of flood.

For the purpose of correlation, the materials thus deposited were separated according to origin and certain topographic and physical characteristics into the following five major divisions: (1) Mississippi alluvium, first bottom soils; (2) Mississippi alluvium, terrace soils; (3) Mississippi-Red River sediments; (4) Red River sediments; and (5) Coastal Prairie sediments of the Gulf Coastal Plain.

These five groups of soils have been again divided into several soil types and named as follows: Group (1) Yazoo, Sharkey and Muck, (2) Lintonia and Olivier, (3) Franklin, (4) Yahola, and (5) Iberia. These differ in pH, movement of water, chemical composition, fertilizer requirements and physical characteristics.

Through the study of these soils in detail, as shown in the paper referred to above, fertilizer experiments have been installed and better fertilization policies have resulted. The newer canes have shown a greater response to fertilization and with the increased yields per acre it has become almost a universal practice to fertilize ratoon crops with nitrogen fertilizers—mostly nitrate of soda and calcium cyanamid. In the coastal plain soils, which are alluvial soils laid down under salty water conditions, somewhat similar to those at Kekaha, there is a distinct response to potash. In the more acid soils (4. — 6. pH) of the older types, responses to both phosphates and potash have been noted. The amounts of plant food used are very small when

compared with Hawaiian practices but are quite reasonable when considered in connection with the very few months of growing weather available.

As mentioned above, the control of cane diseases, and more especially mosaic, is of primary importance to the industry. The exceedingly interesting research carried out on the various strains of mosaic, their different effects on varieties, their effect on each other and their control, is probably one of the most outstanding things seen and discussed at the congress. The control of root rot, of red rot and the research on windrowing losses are all excellent accomplishments shown us while at the Houma Station of the United States Department of Agriculture. Varieties are introduced from the Canal Point Station in Florida and the remarkable accomplishments of these newer canes speak very highly of the work carried on by Dr. E. W. Brandes and his associates in the Sugar Plant Investigations of the Bureau of Plant Industry, U.S.D.A.

The crop is harvested by hand, either by day work, by the ton, or by the "running acre." The day work rate as set by the A.A.A. is \$1.50 per male and \$1.20 per female. The per ton rate is set at 75 cents by the A.A.A. In the other method, or task work, the rate varies with yield and variety but it is set for a specified length of row about 210 feet in length. Thirty-five rows six feet apart and about 210 feet long make an equal-sided acre, and each line is called a "running acre." The rates vary from 45-65 cents per "running acre." By the task method, many good negro cutters make around \$2.00 a day, or slightly more, cutting from 3½-4 tons of cane. Much less is cut by the other methods. Cutting costs are very much higher in Louisiana than in Hawaii. However, the reverse is the case in loading costs.

The most universal method of loading is by the Castaganos, or similar, loaders into wagons. A double self-tripping chain sling is laid in each wagon and the grab loader gathers up bundles of cane weighing a hundred to two hundred pounds and lifts them into the wagon. When there are about 3 tons in the wagon, the chains are fastened and the wagon hauled to a loading station. At the loading station, these slings are lifted out and the large bundle is bound more tightly. Several of these large bundles (about 3 tons each) are loaded onto a trailer or truck and hauled to the mill. At the mill yard, huge crane unloaders lift these bundles onto a pile or onto the unloading platforms next to the carrier. At this point, the chains are removed by tripping the sling. Sufficient cane is piled at the mill during the day to run the factory at night. Huge piles from 30-60 feet in height adjoin the carrier and cover considerable area. The first impression one gets of this procedure is that the cane must become too old in these piles. We were assured, however, that these were cleaned up every 48 hours at the latest which hardly seemed believable. The huge cranes have grabs similar to our harvesting grabs for taking the cane over onto the carrier platform. The costs of loading the cane and delivering it to the main line railroad loading stations do not exceed 30 cents per ton and more often are nearer 20 cents than 30. The railroading to the mill is expensive and usually adds another 20-30 cents per ton, making the total harvesting cost per ton delivered to the mill around \$1.25. Where the trailers deliver their loads of 5 to 7 bundles (14-18 tons) to the mill yard, the costs of harvesting per ton are lower. This usually happens for areas within driving distance of the factory. These trailer trucks are usually of the semi-trailer type with Ford or Chevrolet hauling units. The trailers constructed by several local concerns are of excellent construction and stand up well. These trailers

cost in the neighborhood of \$500.00 and with an \$850.00-\$900.00 truck-hauling unit the total cost is under \$1,400.00. As mentioned above, these readily haul from 14-18 tons per load and travel at a fairly fast rate of speed. These semi-trailers usually have about the following dimensions—27' x 8' x 5'. Some of the best that we saw were made by the Thompson Machinery Company at Labadeville and should make an excellent piece of equipment for any of our plantations wishing to haul cane on their more level fields. Hydraulic brakes may be installed on them for a small amount extra and the trailers used on heavy grades.

The Castaganos loader, and others of a similar type, are motor driven but are usually hauled around with a team of mules. A small tractor (either Allis-Chalmers or Farmall) will haul 2-3 rubber-tired wagons (2 wheel) and the loader will keep three or more of these "trains" going. At one place we noted a loader with its operator, and a driver for the 4 mules, working with 3 tractors (3 drivers) and a ground crew of 4, with 1 man on the wagon being loaded, or a total of 10 men loading about 400 tons a day. This would compare very favorably with our own operations if all track laying, hauling and loaders were included in arriving at an average tonnage per man. The rubber-tired, all steel wagons sell for from \$300.00-\$400.00 and weigh a little less than 1 ton.

After harvesting is completed, the trash is burnt off and the field left to ratoon. Sometimes the soil is cultivated but not as a regular practice until the following spring. Where cane is windrowed before being loaded, the leaves are left on and the whole covered over. Later these canes are dug out, topped and cleaned for harvesting operations. A very novel and successful machine was in operation doing this particular job. There were many points about it that might be used in a cane harvesting machine.

Another interesting machine was the Wurtele cane harvester which was doing a fair job of cutting, topping, and bundling long stalks of cane without stripping off much of the dead leaves. It was working successfully in a 30-ton crop of standing cane.

Milling operations were not often observed by the field group but an opportunity was given the writer to see the Munson cleaner in operation and it seemed to be doing a fair job. The feeding of the short pieces into the crusher seemed to be difficult.

Now an attempt will be made to mention the more outstanding papers presented and consider the points that would be of practical value to Hawaii. Since there were over 135 papers presented to the congress, it will be next to impossible to discuss more than just a small share of them. The twelve delegates from Hawaii divided themselves among the various sections so that someone was present at almost all the sessions held. The papers were divided into the following sections for discussion: pathology, varieties, field and soils, entomology, technique of field experiments, and milling. The writer attended mostly the discussions on field and soil problems, with an occasional visit to those on pathology, experimental technique, and varieties.

In the papers on pathological problems, those on mosaic certainly commanded the most interest. As mentioned above, the work on the various strains of mosaic has been so well carried out that it makes one realize how complicated a study of such a cane disease may become. The mild strains of mosaic secured from Hawaii

were shown to be very much less harmful than those in Louisiana. This is an excellent example of how necessary are the quarantine measures we are maintaining in Hawaii. Healthy seed and roguing are still the best control measures. An interesting paper was presented that tended to show that soils or soil treatments had a marked effect on the severity of mosaic. The inoculating of young seedlings with the various strains of mosaic for determination of immunity and resistance is a very much worth while adjunct to cane breeding operations. Papers were presented to show similar testing for resistance to other cane diseases in seedling propagation work. The work in the Philippines on the transmission of Fiji disease by all the stages of the cane leafhopper makes us think all the more of the troubles we are threatened with when plane service is inaugurated to the South Seas. An interesting abstract on *Pythium* is inserted here.

In a series of greenhouse and field experiments carried out during 1936-38 at the Louisiana Experiment Station at Baton Rouge, Louisiana, the development of corn and sugar cane plants was followed in different soil types, at different pH levels, at different temperatures and under different nutrient conditions with particular reference to the effect of these variations upon the action of *Pythium* organisms attacking the roots and upon the action of *Trichoderma* organisms attacking the *Pythium*.

Under the conditions of the experiments, low temperatures favored the destructive action of the *Pythium*. Modifications of the pH level were without appreciable effect. The application of nitrates was accomplished by an increase in root rot. Treatments with high phosphates were accompanied by a reduction of root rot. In general, however, neither the nitrates nor the phosphates appeared to have any pronounced effect upon the demonstrated antibiotic action of *Trichoderma* against *Pythium*. A strain of *Trichoderma* obtained outside the state proved more effective against *Pythium* than most of the strains obtained locally from sugar cane soils.

The control of seed piece rot and the stimulation in germination from using such materials as Ceresan were discussed in a paper from South Africa and the results compare favorably with those secured on Kauai.

Other pathological papers were presented giving general resumé's for different countries, and discussions on such diseases as sclerotic disease of sugar cane in Formosa, red stripe in Brazil, red rot in Louisiana, gumming in Queensland and Mauritius, and cytospora rot, stubble deterioration, and rhizoctonia in Louisiana.

The papers on varieties were not as numerous as in the past, as many of the detailed discussions on breeding and selection have been so thoroughly covered in past proceedings. Those of more importance were on the subject of breeding for disease resistance and especially that for mosaic and gumming which were mentioned above. A few on taxonomic, and other characteristics, were presented. Most of those presented from Louisiana clearly showed the exceedingly great importance breeding has been to the return of their local industry to a place in the sun. A most interesting paper by Bell of Queensland showed that "the making of second selections of seedlings on the basis of comparative vigor of 6-10 stool plantings from original seedlings was fundamentally unsound." A paper was presented and many pictures were shown of the crosses between sugar cane and bamboo in India. In the writer's estimation, the evidence is not sufficiently strong to prove that such crosses have been culminated.

In the section on field and soil problems, the two matters will be discussed separately. A paper presented by representatives of the A.A.A. on sugar control programs and their effect on the Louisiana sugar industry showed how serious

these measures may really become. In the discussion following the paper very pertinent questions were not answered by the authors and thus the opportunity for a lively discussion fell by the wayside. A few general papers were presented on the following points: interplanting sugar cane to paddy rice; cane cultivation in Mauritius; flood following sugar cane soils; double versus single planting; some aspects of drought resistance; cultivation by rubber-tired tractors; windrowing; transportation; deep tillage work with gyrolettes; drainage of peat soils; and mechanical harvesting in Hawaii. One paper on "Resistance to inversion of sucrose in harvested sugar cane in Louisiana" by Lauritzen and co-workers at the Houma Station was most interesting and the abstract is presented herewith:

The inversion of sucrose in harvested sugarcane in Louisiana is influenced by water content and change in water content resulting from prevailing environmental conditions such as air temperature, relative humidity, and air movement and by such factors as temperature, nutrition, maturity, invertase activity, variety of cane and some unknown factors.

All varieties are resistant to inversion under certain environmental conditions and some varieties under environmental conditions that are normally favorable to inversion.

The varietal response to environmental conditions favorable to inversion is characteristic and constitutional.

The response in certain varieties is not correlated with the invertase content of the stalk and must involve in certain resistant varieties some inhibiting factor independent of the invertase content.

In a given variety grown under given conditions the sucrose content of the stalk is an indication of maturity and thus may be an indication of resistance or susceptibility to inversion. In cane grown under different conditions sucrose content may not be an indication of resistance or susceptibility. Between varieties sucrose content is not an indication of resistance or susceptibility.

There are indications that the varietal characteristics relating to inversion are transmitted from parents to offspring.

The development of resistance to inversion of sucrose in sugarcane is of immense economic importance to the sugar industry wherever sugarcane is grown.

A paper on the "Behavior of Sugarcane to Length of Day" was mostly in connection with flowering and should be of some interest to cane breeders. The windrowing machine by Munson, discussed in the first part of this report and in one of the papers, should materially aid the industry in lowering harvesting costs. Most all windrowing starts on November 15th, and a man may windrow from  $\frac{1}{4}$ - $\frac{1}{2}$  an acre per day by hand. The Munson machine will cover considerably greater area per man per day. Cane often remains in this windrow for 50 days. The other method of research towards reducing costs and losses in windrowing is the breeding of canes better able to stand this severe treatment. Co. 281 is still the best cane for this operation but since it is being so severely damaged by mosaic greater efforts are being made to find something better.

Four excellent papers on the soil problems and types of Louisiana were presented and made use of by the writer in describing the soils in the first part of this report. These showed the variations in physical characteristics and chemical analyses between the several soil types. Considerable variations were noted in attempts to correlate soil analyses with response to fertilizer treatment and to analyses of cane juices. In some soils correlations were good, while others showed none at all.

One paper presented from South Africa gave criticisms of the Hawaiian rapid chemical methods of analyses. Excellent papers from Queensland by Kerr and von

Stieglitz, together with those mentioned above and with comments made by delegates present at the discussions, very clearly illustrated the diversity of problems and conditions in different cane growing countries. The writer pointed out at the meeting that where methods were satisfactory and correlations were secured in any one country it was asking too much to expect these methods and correlations to be true in another country. The important point brought out was that if any methods were helpful to our better understanding of proper nutrition it was of little, if any, importance if these same methods were unsuccessful in another country. As pointed out by one of the most able investigators present at the meeting, it was much more important to put your facts into practical application on your own soils than to rush into print so someone else might find fault with your methods. But, above all, he added: "Be sure that you yourself are sufficiently critical of your own methods so that there is no self-deception."

A paper on water culture work with phosphate deficiency in Formosa presented some interesting facts. References were made to other papers published by the same authors—Saito and Kenjo—on nitrogen and potash deficiencies, and these should be secured and studied together before comments can be made. The writer would like to suggest that these be reviewed and discussed in *The Hawaiian Planters' Record* or the *Director's Monthly Letter* and compared with Hawaiian results.

In the experimental technique section much was said on sampling both cane and soil for representative data. Once again the methods satisfactory for one country may not be that for another. The portable weighing apparatus used in experiments in Louisiana warrants a trial in Hawaii and especially so since size of plot has nearly always been related to the harvesting equipment and the ability to weigh all the cane grown.

The Report of the Committee on Technique of Field Experiments together with the papers by Borden; Kerr; Arceneaux, Belcher, Gibbens and Stokes; Holmes and O'Neal; and Williams and Follett-Smith, all brought out the differences of opinion on experimental layout and interpretation of results. Some of these seemed to be giving too much consideration to the actual data secured and not sufficient to the practical considerations of the things that may have happened to produce those particular results. On the whole, these papers are of considerable interest and worth greater study and review by someone more versed in their details than the writer of this report.

The entomological papers and discussions will be more ably handled by C. E. Pemberton and those on milling by S. S. Peck.

A most interesting abstract of the paper on "The Cell-sap concentration of sugar cane varieties in relation to their resistance to the attack of white leaf louse" by Yamasaki and Arikado, is presented herewith:

Tests of the cell-sap concentrations in the leaves of twenty varieties of sugar cane, half of them resistant and half of them susceptible to the attack of the white leaf louse, disclosed the fact that all of the cell-sap concentrations in the susceptible canes were appreciably higher than in the resistant canes. Further tests of infested and non-infested leaves gave similar differences. The minimum critical concentration at which cane leaves appeared subject to attack was about 4.5 (Ref. Brix). In addition to varietal differences in cell-sap concentration, it was observed that the addition of water by rain or irrigation reduced the cell-sap concentration. The results suggest possible control by choice of varieties and irrigation where indicated, cell-sap concentrations being taken as indices.

In the milling section, the following two papers seemed of particular interest to the writer: "The use of phosphoric acid in cane sugar manufacture and refining" by George P. Meade, Manager of Gramercy Refinery of the Colonial Sugars Company, and "Developments of J. J. Munson for improving conditions of sugar mill operations" by E. L. Denis, Consulting Engineer of Godchaux Sugars, Inc.

On the whole, the meeting and tour did much to stimulate our interest in many ways but mostly in regard to harvesting equipment and disease control. It is hoped that more members of our local society will become members of the International one and secure the printed proceedings for a more thorough study. The writer wishes to thank you for the privilege of presenting this report and hopes there may be something of interest in it. However, he wishes to clearly point out that the above figures and comments on operations in the Louisiana cane belt are subject to criticism. It hardly seems possible that an absolutely accurate picture could be secured in so short a visit. Apologies are offered for the mistakes, if they exist, and to anyone who may have been hurt by the writer's comments.

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## Sugar Prices

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96° CENTRIFUGALS FOR THE PERIOD  
SEPTEMBER 26, 1938 TO DECEMBER 5, 1938

Date	Per pound	Per ton	Remarks
Sept. 26, 1938..	3.05¢	\$61.00	Cubas; Philippines.
“ 30.....	2.99	59.80	Philippines.
Oct. 5.....	3.02	60.40	Philippines; Puerto Ricos.
“ 6.....	3.0833	61.67	Puerto Ricos, 3.08; Philippines, 3.10; Cubas, 3.10, 3.07.
“ 7.....	3.135	62.70	Philippines, 3.15; Cubas, 3.12.
“ 14.....	3.10	62.00	Cubas.
“ 18.....	3.05	61.00	Cubas.
“ 21.....	3.10	62.00	Cubas.
Nov. 1.....	3.08	61.60	Cubas.
“ 3.....	3.05	61.00	Cubas; Philippines.
“ 22.....	3.00	60.00	Puerto Ricos.
Dec. 5.....	2.85	57.00	Cubas.

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# THE HAWAIIAN PLANTERS' RECORD

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## **In This Issue:**

### *A Modern Statistical Analysis for Field Experiments:*

With a new interest being shown in the modern methods of studying the results from field experiments, it seems opportune to present a non-technical discussion, with working examples, of the "analysis of variance"—a statistical tool which may be used to measure the significance of the effects of the applied treatments. This is especially necessary since our attention has become focused on the plan of field testing in "factorial experiments," for it has made possible a reliable estimate of the results obtained from these more complex plans.

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### *Pythium Root Rot of Sugar Cane in Louisiana:*

Exhaustive studies of *Pythium* root rot of sugar cane in Louisiana during the period since 1924 are reported by R. D. Rands and Ernest Dopp in Technical Bulletin 666 of the U. S. Department of Agriculture, published in October 1938. This valuable contribution to an understanding of the root rot problem is reviewed briefly and the authors' summary is quoted in full, because of the importance of this disease to our industry when varieties are naturally susceptible or become so in a modified soil environment.

The failure of the old noble varieties in 1923-1926, in part due to *Pythium* root rot, caused a virtual collapse of the Louisiana sugar industry. Varietal introductions by the U. S. Department and cooperating agencies restored the industry to the prosperity level existing prior to 1907. Root rot is reported as being the most important factor in the root disease complex which continues to be a serious problem on these more resistant and vigorous hybrid canes.

The responsible agents in root rot are fungi of the genus *Pythium*, of which *Pythium arrhenomanes* is by far the most active. This species is identical with the *Pythium* sp. first reported in Hawaii as a cause of root rot of cane in 1919. The reported increase in virulence of *P. arrhenomanes* coincident with the general adop-

tion of resistant varieties which favored the survival of the more virulent strains, indicates that root rot is a dynamic rather than a static factor in cane culture.

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#### *Influence of Potash Fertilization Upon the Production and Composition of Dry Matter:*

Studies that show effects which the stage of maturity of a crop may have upon its composition, and also the lack of a correlation between yields and their potash content, make it appear improbable that a recommendation for specific potash fertilization can safely be based upon the crop's potash-composition figures.

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#### *The Growth of Plants in Water and Sand Cultures:*

Soiless agriculture and its synonyms, plant requirements, the development and use of a soluble plant fertilizer, the water- versus the sand-culture method for growing plants, and practical uses of soiless agriculture are discussed in this article. The preparation of two complete nutrient solutions and directions for their use by Hoagland and Arnon of the University of California are given for the benefit of those who may wish to prepare their own culture solutions.

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#### *Variation in Available Nutrients in an Uncropped Surface Soil:*

A semi-monthly analysis, for available nutrients from an uncropped soil during a period of two years, reveals variations which indicate the limitation of soil analyses and the hazard of a strictly quantitative interpretation therefrom.

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#### *Colorimetric Method for the Determination of Sulfate in Cane Juice:*

A colorimetric method for determining the amount of sulfate in cane juice is presented. The method is rapid, especially when using the permanent inorganic color standards which are described. The sulfate in the juice, which is brought to a degree of acidity represented by a pH of about 4.0, is precipitated with an excess of standard barium chloride solution. Sodium rhodizonate is added to the excess barium which forms the colored solution and the latter is compared with the standards. A sample of crusher juice can usually be analyzed to within a few per cent of its actual sulfate content in a matter of minutes. The method is applicable to other aqueous solutions containing sulfates.

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#### *The Third Study of Water and Cane Ripening:*

When the amount of water in the soil reaches the wilting point, very little production of sugar can take place in the leaves of the sugar cane plant. A plentiful supply of water is essential not only for the formation of sucrose in the blades, but also to facilitate its transport to the stem and its expression in the juice.

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# A Modern Statistical Analysis for Field Experiments

## The Analysis of Variance for Simple Factorial Experiments

BY R. J. BORDEN

### INTRODUCTION

The application of statistical measures to results from field experiments provides a safety factor which discourages attempts to definitely attribute small apparent differences, which have not been adequately measured, to the known differential treatments which have been applied. Through their use we are also able to determine the limits between which the true value of a treatment effect is most likely to occur.

The field investigator of sugar cane problems should look upon a statistical analysis simply as another technical tool for his use. It will take its proper place along with his surveying instruments, his steel tape, scales, refractometer, calculating machine, slide rule, and mathematical tables and formulae, in helping him to note and measure the factors and relationships that are involved in his investigation. It does not, however, furnish the explanation of these observations. Neither does it supply a substitute for clear-cut thinking, nor for his logical common-sense analysis that is based on knowledge he has secured from past experience, nor can it compensate in any way for an unskilled or an inadequate technique which may have been used.

When a statistical analysis has been completed, its results must be compared with the original assumptions which were set up when the objective of the investigation was proposed as a field test problem. If the analysis agrees with the assumptions, the results are clearly interpreted and should then be stated in terms which are understandable to those who will use them. If the statistical result and the hypothesis are not in agreement, both will need to be carefully re-examined—the hypothesis, to see if it was correctly deduced; the statistics, to see if they were properly and correctly worked out. Then, if the hypothesis and the results are still found to be inconsistent, the results will need to be recorded as actual observations that are not consistent with the expected results and hence subject to further study before definite conclusion is drawn.

Agronomists are in general agreement that a minimum level for judging whether measured differences in field experiments are most likely the effect of chance, (and hence not definitely attributable to known treatment differentials), is represented by a probability of 5 in 100 ( $P = .05$ ), more commonly spoken of as odds of 19 to 1. If we use this 1-in-20 level of significance in judging our results, we must recognize that 1 out of every 20 results may be judged significant when there are no treatment effects at all. Hence we must avoid emphasis on such statistical significance when the results apparently have no consistent relationship with a well-established belief based on long experience: judgment had best be reserved until further evidence is secured.

An interpretation of non-significance does not mean that no treatment effect exists; it simply means that the observed apparent effect would be likely to show

up in excess of once in twenty results by chance alone, even if there were no real treatment effects.

Plantation men who are interested in conducting field experiments, as well as those who study their results, are being impressed with the scope and values which the modern statistical methods offer. All too few, however, who have occasion to use statistical methods are sufficiently mathematically inclined to think in mathematical terms, and most of those who wish to interpret the data themselves are inclined to look askance at all mathematical expressions and symbols. Hence, in the belief, based on personal experience, that the application of statistical measures to the results of field experiments can best be learned by actual practice in their application, we have prepared the examples which follow with complete work sheets so that he who is interested may follow the mechanics of the mathematical procedures which are used.

It is more than likely that these plans which we have discussed in the examples that follow will be criticized by the strict adherents to a policy of randomization for plot positions in experimental areas. Better men than we have argued the pros and cons of randomization. For our part, we have convinced ourselves that more reliable yields from plots carrying a mature sugar cane crop can be secured when such plots have had an intelligent arrangement than when they have been assigned their position through randomization. "Student,"\* after looking over our designs for balanced arrangements and the results of many studies we have made from our Blank Test data, once wrote us that "your results are just what I should have expected, that is to say, that the balanced arrangement is better than the random," and again, "I am quite sure that there is much to be gained by a balanced arrangement."

There are few who question our contention that the cane yields will be more reliable from a balanced than from a random plot arrangement, but there are many who contend that no valid estimate of error can be made for such yields because the plot arrangement has not been at random. Student has commented that "all these people mean by valid is that they are rigidly justified in using the various (statistical) tables, and that is an advantage which is not only not worth going far to get but may often be dearly bought. . . . What is claimed, is that if a randomized layout is used, the various tables, Z, T, etc., will give you the probability of obtaining eccentric results, but this is only true *before* the actual layout is selected from among the possible random arrangements. *After* the particular layout is chosen, the tables no longer apply strictly, and this can be seen from the fact that the regular arrangements are among those which could be drawn at random. It is not, unfortunately, possible to estimate the probability accurately in any but a random arrangement, but that does not prevent your being able to say that a balanced arrangement, for example, will give results which are more accurate than those which would have been obtained with a random arrangement."

The fact, as Student points out, that a balanced arrangement *may* actually have been drawn at random, and that if it had been so drawn, one would be correct in using the statistical tables and methods which would be denied if the balanced arrangement was deliberately planned, fails to make common sense for most agronomists.\* The further fact, that this tossing of a coin or drawing from a hat to decide the plot arrangement, may actually place several plots of the same treat-

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\* The late W. S. Gosset in personal communication.

ment adjacent and coinciding with an abnormally high-or-low-fertility spot, or at such a position within the field test area where one knows before the test is started that a biased result will be secured, does not elicit much support from a practical field man who knows the conditions under which field testing must be done. Furthermore, the fact that most agronomists have quite arbitrarily set odds of 19 to 1 ( $P=.05$ ) as their minimum level of significance makes it appear unreasonable to insist on a blind and rigid adherence to precise mathematics, for we would accept odds of 20 to 1 as significant and consider odds of 18 to 1 as not significant in our final interpretation. Hence, unless our experimental plans will give us yields with as little error as possible, that is, yields which are free from bias, then any measure of significance is apt to give a false degree of confidence in the results. Surely then it is more reasonable to err, if by chance we err at all, in the direction of *estimating* the significance of a result than in actually obtaining such result; the really important large differences between field treatments will be detected without a highly precise mathematical tool.

We might argue this issue still further but we have probably said enough to indicate that we feel satisfied that the ordinary statistical tables can be intelligently used to study data from balanced arrangements. The required randomness for plot positions is most likely secured when the decision is made to locate the first plot of a plan, which has been drawn up in the office, at a certain point in the field, since the heterogeneous nature of soil fertility in our cane lands provides thereafter the important features of a random distribution.

#### THE ANALYSIS OF VARIANCE

Although R. A. Fisher, of Rothamsted, introduced his method of analysis of variance some 15 years ago, it has only been in more recent years that our American agronomists have quite generally adopted it for studying the results of their experiments.

As applied to field experiments, the analysis of variance first separates the total variation of the experiment into two parts—one identified with the differences between the plot yields which have received the same treatment (variation *within* treatments), and the other identified with the variation among the means of the different treatments (variation *between* treatments). After this separation has been made, we are able to test whether the treatment means are significantly different, that is, whether they vary more than would be expected by chance alone. For example, we may take the following yields (T.C.A.) from 4 replications of each of 3 treatments tested on 12 plots:

T.C.A. FROM 3 TREATMENTS (A, B, X)				
	A	B	X	Total
	62	64	54	
	68	60	60	
	62	68	52	
	66	72	50	
	<hr/>	<hr/>	<hr/>	<hr/>
Sums	258	264	216	738
Means	64.5	66.0	54.0	61.5

The total variation in this experiment may be represented by the sum of all of the squared deviations ( $d$ )<sup>2</sup> of each plot yield from the mean yield of all plots (61.5), e.g.

A		B		X		Total variation
d	d <sup>2</sup>	d	d <sup>2</sup>	d	d <sup>2</sup>	
+ .5	.25	+ 2.5	6.25	— 7.5	56.25	
+ 6.5	42.25	— 1.5	2.25	— 1.5	2.25	
+ .5	.25	+ 6.5	42.25	— 9.5	90.25	
+ 4.5	20.25	+ 10.5	110.25	— 11.5	132.25	
						<hr/> 505.00

Note: The sum of the + and — figures in the "d" columns should equal zero (in this example, we have + 31.5 and — 31.5).

The amount of variation, which exists between the plot yields which have received the same treatment, is then found by summing the squared deviations of each plot yield from the mean yield of its respective treatment ("A" at 64.5, "B" at 66.0; "X" at 54.0), e.g.

A		B		X		Total variation within treatments
d	d <sup>2</sup>	d	d <sup>2</sup>	d	d <sup>2</sup>	
— 2.5	6.25	— 2.0	4.0	0	0	
+ 3.5	12.25	— 6.0	36.0	+ 6.0	36.0	
— 2.5	6.25	+ 2.0	4.0	— 2.0	4.0	
+ 1.5	2.25	+ 6.0	36.0	— 4.0	16.0	
						<hr/> 163.00

This pooled sum of the squared deviations (163) represents an uncontrolled variation *within* all of the treatments (often called the experimental error) and when subtracted from the total sum of the squares (505) it gives the sum of the squares which indicates the variation *between* the treatments: 505 — 163 or 342. This sum of the squares *between* treatments (342) may also be secured by squaring the deviation of each treatment mean from the general mean (61.5), summing these squared deviations, and multiplying this sum by 4 (since there are 4 plots represented in each treatment mean), e.g.

Treatment		Mean yield	d	d <sup>2</sup>
A	=	64.5	+ 3.0	9.00
B	=	66.0	+ 4.5	20.25
X	=	54.0	— 7.5	56.25
<hr/>				
General mean		61.5		85.50
				× 4
				<hr/> 342

Any sum of squares divided by its respective number of degrees of freedom\* gives the variance. The degrees of freedom for our example are as follows:

Between treatments .....	2
Within treatments (3+3+3).....	9
<hr/>	
Total .....	11

\* Degrees of freedom: Equivalent to  $n - 1$ , i.e., one less than the number of observations concerned in the particular sum-squares calculation.

Thus the total variance would be  $505 \div 11$  or 45.9, and the variance *between* treatments (i.e., due to the treatment differences) is  $342 \div 2$  or 171. Similarly the variance *within* treatments (i.e., due to position and to the unidentified error) is  $163 \div 9$  or 18.1.

However, it is upon the ratio of the *mean* variance *between* treatments to the *mean* variance *within* treatments that the test of significance is based. Snedecor has given us mathematical tables for determining whether this ratio,  $\frac{\text{mean variance between treatments}}{\text{mean variance within treatments}}$  which he calls "F," is significant.

So we set up the foregoing facts for the analysis of variance as follows:

Source of variation	Degrees of freedom	Sum of squares	Mean square or variance	"F"	
				Found(1)	Required(2)
Between treatments	2	342	171	9.4	4.26
Within treatments	9	163	18.1		
Total	11	505			

$$^{(1)} \frac{171.0}{18.1} = 9.4$$

<sup>(2)</sup> A figure obtained from Snedecor's tables, for a probability of .05 or odds of 19 to 1, and the proper number of degrees of freedom.

Therefore, since "F" as found is greater than "F" required, this analysis would indicate that the mean variance between or for treatments is significant, i.e., it would not be expected to occur by chance more often than once in twenty similar tests.

Mathematical calculations concerned with analysis of variance may be greatly simplified by using a shortened method and working with totals rather than averages, and hereafter in our examples we shall use the short methods. The procedure used in the short method for finding the sum squares from the example just cited would be as follows (using same data):

A		B		X		Total Y <sup>2</sup> (all treatments)
Y	Y <sup>2</sup>	Y	Y <sup>2</sup>	Y	Y <sup>2</sup>	
62	3844	64	4096	54	2916	
68	4624	60	3600	60	3600	
62	3844	68	4624	52	2704	
66	4356	72	5184	50	2500	
Sums	258	264		216		45892
Total (T) 258 + 264 + 216 = 738.						

$$\text{Correction factor} = \frac{T^2}{n} = \frac{(738)^2}{12} = \frac{544644}{12} = 45387$$

Total Sum-Squares:\*

$$\begin{array}{r} 45892 \\ -45387 \\ \hline \end{array}$$

Corrected Total Sum-Squares 505 (with 11 degrees of freedom)

\* This term "Sum-Squares" always refers to the sum of the squares of the deviations from the mean yield of all plots.



*Sum-Squares Treatments:*

$$\frac{(258)^2 + (264)^2 + (216)^2}{4} = \frac{182916}{4} = 45729$$

Corrected Sum-Squares Treatments . 342 (with 2 degrees of freedom)

*Sum-Squares Error\** (i.e., within treatments):

$505 - 342 = 163$  (with 9 degrees of freedom)

These figures are then set up for further examination in the Analysis of Variance Table as previously shown on page 77.

To better understand the “Analysis of Variance,” one needs to be aware of the following relationships:

1. That the Total Sum-Squares is made up of the Sum-Squares *between* the Treatments and the Sum-Squares *within* the Treatments; furthermore, that the Sum-Squares *within* Treatments is made up of the Sum-Squares due to Position (Blocks, Rows and Columns, Squares) and to the Sum-Squares due to Error. (In complex or factorial experiments it will also include the sum of the squares due to the interactions of the various factors.)

2. That the Total Sum-Squares for an experiment is likewise made up of the Sum-Squares *between* the Blocks (or Rows and Columns in a Latin-square layout) and the Sum-Squares *within* the Blocks; furthermore, that the Sum-Squares within Blocks is made up of the Sum-Squares for Treatments and for Error.

3. That the Sum-Squares for position may be identified and determined, (a) for Blocks (in a block arrangement); (b) for Rows and Columns (in a single Latin square); and (c) for Rows, Columns, and Squares (in a multiple Latin-square layout).

4. That the Sum-Squares for the combined Treatments in a factorial experiment (which we shall discuss later) is made up of the Sum-Squares for each of the separate factors and the Sum-Squares of their Interactions.

5. That the respective mean Variances are found by dividing their Sum-Squares by their number of degrees of freedom ( $n - 1$ ).

6. That the measures of significance (odds, "F" values, etc.) are based on the relation between the mean variance *between* Treatments and the mean variance *within* Treatments, i.e.,  $\frac{\text{Mean variance between treatments}}{\text{Mean variance within treatments}}$  = measure of significance.

If the extent of certain contributing factors to the variation *within* treatments can be identified, its magnitude can be reduced and the statistical measurement becomes more refined. Thus, when certain layouts or arrangements of plots have been used, a positional effect (from blocks, or from rows and columns, or from squares) can be determined and subtracted; in factorial experiments when treatments have been combined, an effect of interaction can be measured and deducted from this variation within the treatments. After such deduction, the unidentified

\* The Sum-Squares within the Treatments is convertible into the Sum-Squares for Error if its component parts are not still further identified.

remainder is the only part which is due to error, and our fraction or ratio for measuring the effect of treatment then becomes  $\frac{\text{Variance between treatment}}{\text{Variance for error}}$

For example, we might have these data from a test on 30 plots carrying 10 replicates of 3 treatments:

Total Sum-Squares for 30 plots = 400, with 29 degrees of freedom.  
 Sum-Squares between 3 treatments = 60, with 2 degrees of freedom.  
 Sum-Squares within 3 treatments = 340, with 27 degrees of freedom.

The mean variance for treatments is then  $\frac{60}{2}$  or 30, while the mean variance within Treatments (the error) is  $\frac{340}{27}$  or 12.6.

The "F" value found would then be  $\frac{30}{12.6}$  or 2.4 and as this is less than "F" required (3.35) for minimum significance, we would have to conclude that the treatment effect may have been due to chance.

On the other hand, if the test has been laid out so that some effect of the positional variation can be determined, and let us assume that we have found a Sum-Squares for 10 Blocks (each Block carrying one plot each of all 3 treatments) amounting to 240, our Sum-Squares within Treatments (340) can be reduced by 240, and the Sum-Squares for Error is thereby only 100; then with 27 minus 9 or 18 degrees of freedom left for Error (because the Blocks have accounted for 9 degrees of freedom), the mean variance for Error becomes  $100 \div 18$  or 5.6. The "F" value now becomes  $\frac{30}{5.6}$  or 5.36, which is more than the "F" required (3.55) for significance, and hence gives a fair degree of assurance that there has been a real effect of treatment.

So let us examine and explain two completed examples, one which illustrates the steps in using this statistical measure on yields obtained from a plot arrangement in Blocks, and the other from an arrangement in a double Latin square:

Our first example is with 2 treatments (A, X), balanced, in 8 blocks (I to VIII) using 16 plots.

Arranged as follows:

YIELDS AS T.C.A.					
I	A	76	X	70	V
	X	74	A	72	
II	X	79	A	77	VI
	A	81	X	83	
III	A	73	X	80	VII
	X	73	A	82	
IV	X	71	A	74	VIII
	A	76	X	70	

DETAIL FOR THE ANALYSIS OF VARIANCE	
Source	Degrees of freedom
Blocks .....	7
Treatments .....	1
Error .....	7
Total .....	15

The *Total Sum of the Squares* of the deviations of each plot yield from the average yield of all the plots includes all of those factors (treatment, position, unknowns) that make one plot yield differently from another.

This *total sum-squares* is most easily obtained by squaring each plot yield, summing these squares, and deducting the proper correction factor (which is the squared sum of all plot yields divided by the number of plots represented in this sum). For example:

$$\begin{array}{rcl} \text{Correction factor } (76 + 74 + 79 + 81 + \dots 82 + 74 + 70)^2 & = & \frac{(1211)^2}{16} = 91658 \\ \text{Then } (76)^2 + (74)^2 + (79)^2 + \dots (82)^2 + (74)^2 + (70)^2 & = & 91931 \\ \text{Minus the correction factor} & & \underline{91658} \\ \text{Corrected Total Sum-Squares} & & 273 \text{ (with 15 degrees of freedom)} \end{array}$$

This *Total Sum-Squares* is made up of the variation *between* the Blocks and the variation *within* the Blocks.

We next obtain the amount of variation which is contributed by the position of the 8 Blocks on the experimental area. Since each Block is similar, in that it contains one plot of both treatments, we determine the Block totals:

Block I	76 + 74 = 150	Block V	70 + 72 = 142
Block II	79 + 81 = 160	Block VI	77 + 83 = 160
Block III	73 + 73 = 146	Block VII	80 + 82 = 162
Block IV	71 + 76 = 147	Block VIII	74 + 70 = 144

These Block totals are then squared, the squared totals summed, and the sum divided by the number of individual plot yields which were represented in each Block total used. The correction factor is then deducted and the result is the amount of the Total Sum-Squares that can be allocated to the Block's position, i.e., the amount between Blocks. For example:

$$\begin{array}{rcl} \frac{(150)^2 + (160)^2 + (146)^2 + \dots (162)^2 + (144)^2}{2} & = & \frac{183769}{2} = 91885 \\ & & \underline{-91658} \\ \text{Corrected Sum-Squares between Blocks} & & 227 \text{ (with 7 degrees of freedom)} \end{array}$$

The difference between the Total Sum-Squares and the Sum-Squares *between* Blocks is the Sum-Squares *within* the Blocks; this would be 273 — 227 or 46. This Sum-Squares *within* Blocks is made up of variation which is due to the different treatments and to the undeterminable effects which we call "Error."

The Sum-Squares for Treatments can be determined by squaring the treatment totals, dividing by the number of plot yields represented in each treatment total, and subtracting the correction factor. For example:

Treatment "A" totals: 76 + 81 + 73 + 76 + 72 + 77 + 82 + 74 = 611 (Avg. 76.4 T.C.A.)  
 Treatment "X" totals: 74 + 79 + 73 + 71 + 70 + 83 + 80 + 70 = 600 (Avg. 75.0 T.C.A.)

$$\begin{array}{rcl} \text{Then: } \frac{(611)^2 + (600)^2}{8} & = & \frac{733521}{8} = 91665 \\ & & \underline{-91658} \end{array}$$

Corrected Sum-Squares for Treatments 7 (with 1 degree of freedom)

The difference between the Sum-Squares within the Blocks and this Sum-Squares for Treatments is the Sum-Squares for Error, upon which our test of significance is to be based. Hence  $46 - 7$  or  $39$  is the Sum-Squares for Error in this experiment (and this item will have the remaining or  $7$  degrees of freedom).

It is now necessary to determine the mean variance for Treatment and for Error, because it is from the ratio of these two mean variance figures that we secure our estimate of the effect of the treatment. These mean variances are determined by dividing the proper Sum-Squares by their respective "degrees of freedom." Thus we have:

For Treatment:  $7 \div 1$  or  $7.0$  as the mean Treatment variance, and

For Error:  $39 \div 7$  or  $5.57$  as the mean Error variance,

and the ratio of Treatment variance to Error variance is  $\frac{7.0}{5.57}$  or  $1.26$ .

Reference to Snedecor's tables for "F" will indicate that this value ( $1.26$ ) is far below the required value ( $5.59$ ) which agronomists will accept as satisfactory evidence of an effect of treatment. Hence the differences found between the Treatment totals are considered as quite apt to be due to chance alone, and we record them as "not significant."

Our second example is with  $3$  treatments (A, B, C) arranged in  $2$  Latin squares, and using  $18$  plots.

Arranged as follows:

	SQUARE I			SQUARE II		
	Column 1	Column 2	Column 3	Column 1	Column 2	Column 3
Row 1	A 80	B 84	C 86	B 84	C 96	A 78
Row 2	C 89	A 84	B 78	A 77	B 83	C 94
Row 3	B 88	C 92	A 76	C 97	A 80	B 89

#### Details of Work Sheet:

SQUARE I			
Row totals	Column totals	Treatment totals	
$80 + 84 + 86 = 250$	$80 + 89 + 88 = 257$	"A" $80 + 84 + 76 = 240$	
$89 + 84 + 78 = 251$	$84 + 84 + 92 = 260$	"B" $84 + 78 + 88 = 250$	
$88 + 92 + 76 = 256$	$86 + 78 + 76 = 240$	"C" $86 + 89 + 92 = 267$	
Square totals 757	757	757	

SQUARE II			
Row totals	Column totals	Treatment totals	
$84 + 96 + 78 = 258$	$84 + 77 + 97 = 258$	"A" $78 + 77 + 80 = 235$	
$77 + 83 + 94 = 254$	$96 + 83 + 80 = 259$	"B" $84 + 83 + 89 = 256$	
$97 + 80 + 89 = 266$	$78 + 94 + 89 = 261$	"C" $96 + 94 + 97 = 287$	
Square totals 778	778	778	

Correction factor:  $\frac{(757)^2}{9} = 63672$  (for Square I only)

Correction factor:  $\frac{(778)^2}{9} = 67254$  (for Square II only)

Correction factor:  $\frac{(757 + 778)^2}{18} = \frac{(1535)^2}{18} = 130901$  (for Totals)

Total Sum-Squares:

$$(80)^2 + (89)^2 + (88)^2 + (84)^2 + \dots (94)^2 + (89)^2 = \begin{array}{r} 131637 \\ -130901 \\ \hline \end{array}$$

Corrected Total Sum-Squares 736 (with 17 degrees of freedom)

Sum-Squares for Rows in Square I:

$$\frac{(250)^2 + (251)^2 + (256)^2}{3} = \frac{191037}{3} = 63679$$

—63672

Corrected 7 (with 2 degrees of freedom)

Sum-Squares for Rows in Square II:

$$\frac{(258)^2 + (254)^2 + (266)^2}{3} = \frac{201836}{3} = 67279$$

—67254

Corrected 25 (with 2 degrees of freedom)

Corrected Sum-Squares for rows in both squares:

$$7 + 25 = 32 \text{ (with } 2 + 2 \text{ or } 4 \text{ degrees of freedom)}$$

Sum-Squares for Columns in Square I:

$$\frac{(257)^2 + (260)^2 + (240)^2}{3} = \frac{191249}{3} = 63750$$

—63672

Corrected 78 (with 2 degrees of freedom)

Sum-Squares for Columns in Square II:

$$\frac{(258)^2 + (259)^2 + (261)^2}{3} = \frac{201766}{3} = 67255$$

—67254

Corrected 1 (with 2 degrees of freedom)

Corrected Sum-Squares for columns in both squares:

$$78 + 1 = 79 \text{ (with } 2 + 2 \text{ or } 4 \text{ degrees of freedom)}$$

Sum-Squares for Two Squares:

$$\frac{(757)^2 + (778)^2}{9} = \frac{1178333}{9} = 130926$$

—130901

Corrected Sum-Squares Squares 25 (with 1 degree of freedom)

Sum-Squares for Treatments and Squares:

$$\frac{(240)^2 + (250)^2 + (267)^2 + (235)^2 + (256)^2 + (287)^2}{3} = \frac{394519}{3} = 131506$$

—130901

Corrected 605 (with 5 degrees of freedom)

*Sum-Squares for Treatments A, B, C:*

$$\frac{(240 + 235)^2 + (250 + 256)^2 + (267 + 287)^2}{6} =$$

$$\frac{(475)^2 + (506)^2 + (554)^2}{6} = \frac{788577}{6} = 131430$$

—130901

Corrected 529 (with 2 degrees of freedom)

*Sum-Squares for Interaction (between Treatments and Squares):*

$$605 - (25 + 529) = 51 \text{ [with } 5 - (1 + 2) \text{ or 2 degrees of freedom]}$$

*Sum-Squares for Error:*

$$736 - (32 + 79 + 25 + 529 + 51) =$$

$$20 \text{ [with } 17 - (4 + 4 + 1 + 2 + 2) \text{ or 4 degrees of freedom]}$$

And from the foregoing, we can set up a table for the analysis of variance, as follows:

Source of variation	Degrees of freedom	Sum of squares	Mean square or variance
Rows .....	(2+2) 4	32	8.0
Columns .....	(2+2) 4	79	19.8
Squares .....	1	25	25.0
Treatments .....	2	529	264.5
Interaction (Treatment and Squares)*..	2	51	25.5
Error .....	4	20	5.0
Total .....	17	736	

Our chief interest is now in the ratio of this treatment variance (264.5) to the error variance (5.0). Since their ratio ("F") of  $\frac{264.5}{5.0}$  equals 52.9 and the required "F" for significance (from Snedecor's tables) is only 6.94, we have a good indication that the treatments have been effective.

Since the analysis has indicated a definite effect of treatment, we can now calculate the significance of the differences between the individual treatment totals.

The standard deviation for the experiment is the square root of the error variance, i.e.,  $\sqrt{5.0}$  or 2.24. The standard error for the total of 6 plots of any one treatment is  $\sqrt{5.0 \times 6}$  or 5.48, and a difference between any two such totals greater than  $t\sqrt{5.0 \times 6 \times 2}$  or  $2.78t\sqrt{60.0}$  which is 21.5 tons is significant. Treatment totals are:

$$\text{"A"} = 475 \text{ tons} \quad \text{"B"} = 506 \text{ tons} \quad \text{"C"} = 554 \text{ tons}$$

\* Note: Sometimes one finds a significant interaction between Treatments and Squares, that is, there may be a difference in the effectiveness of the treatments when tested on different parts of the experimental area. In the present example, this type of interaction is probably not in effect since the ratio of the Interaction variance to the Error variance  $\frac{25.5}{5.0}$  or 5.1 is less than the required "F" (6.94) which is needed for significance, thus indi-

cating that this particular interaction effect might quite easily be due to chance. When such an interaction is not significant, it may be advisable to put its associated sum-squares and degrees of freedom back with error, to afford a more conservative estimate of the treatment effects.

† The values of "t" for the number of degrees of freedom associated with the error variance are obtainable from Fisher's tables of "t" values.

Hence both treatments "B" and "C" are distinctly better than "A," and "C" is also better than "B." If desirable, these treatment totals can now be expressed as averages by dividing by the number of plots represented: thus the "A" treatment average would be 79.2 tons, the "B" average is 84.3 tons, and "C" averages 92.3 tons.

From the foregoing discussions it should be clear just how the precision by which the effect of treatment can be measured is increased when the variation within treatments can be broken down into the effects of position and the effect of error. But this assumes that the plot arrangement has been of such a nature that the amount of this positional variance can be determined. So if we are to use this method of statistical analysis, we must be certain that our experimental layout has been planned to allow for the best identification of its positional variance.

For reference, and as acceptable examples for our "Grade A" sugar cane experiments, we offer the 4 following plans (Figs. 1 to 4).

#### PLANS FOR ANALYSIS OF VARIANCE

For any 3 treatments (A, B, X) using a total of 27 plots

IN MULTIPLE LATIN SQUARES (I II III)

I	A	B	X	A	X	B	III
	B	X	A	X	B	A	
	X	A	B	B	A	X	
II	X	B	A				
	B	A	X				
	A	X	B				

Fig. 1

#### DETAIL FOR THE ANALYSIS OF VARIANCE

Source	Degrees of freedom
Rows (2+2+2) = .....	6
Columns (2+2+2) = ....	6
Squares .....	2
Treatments .....	2
Interaction*	
(Treatments & Squares)	4
Error .....	6
Total .....	26

\* See footnote on page 83.

IN BALANCED ARRANGEMENT IN 9 BLOCKS (I to IX)

V

I	A	X	A	VIII
	B	B	B	
	X	A	X	
II	X	A	X	IX
	B	B	B	
	A	X	A	
III	A	X		
	B	B		
	X	A		
IV	X	VII		
	B			
	A			

Fig. 2

#### DETAIL FOR THE ANALYSIS OF VARIANCE

Source	Degrees of freedom
Blocks .....	8
Treatments .....	2
Error .....	16
Total .....	26

For any 4 Treatments (A, B, C, D) using a total of 32 plots

IN DOUBLE LATIN-SQUARE  
ARRANGEMENT

I	A	D	B	C
	B	C	A	D
	C	B	D	A
	D	A	C	B
II	D	B	C	A
	A	C	B	D
	B	D	A	C
	C	A	D	B

Fig. 3

DETAIL FOR THE  
ANALYSIS OF VARIANCE

Source	Degrees of freedom
Rows (3+3) .....	6
Columns (3+3) .....	6
Squares .....	1
Treatments .....	3
Interaction (Squares & Treatments)	3
Error .....	12
Total .....	31

IN BALANCED ARRANGEMENT  
IN 8 BLOCKS

I	IV			VII
	A	D	A	
	B	C	B	
	C	B	C	
II	D	A	D	VIII
	C	B	C	
	B	C	B	
	A	D	A	
III	VI			
	A	D		
	B	C		
	C	B		
	D	A		

Fig. 4

DETAIL FOR THE  
ANALYSIS OF VARIANCE

Source	Degrees of freedom
Blocks .....	7
Treatments .....	3
Error .....	21
Total .....	31

FACTORIAL EXPERIMENTS FOR SUGAR CANE

Ordinarily it has been our custom to install separate experiments to test separate issues. Thus we have had our "Amounts-of-Nitrogen" (AN) tests, and our "Amounts-of-Potash" (AK) tests; our "Variety" tests, and our "Cultivation" tests. In these AN tests, we have arbitrarily set a uniform total for the potash (and phosphate) applications at what we *believe* to be an adequate amount to insure its not being another limiting factor; in the AK tests we have specified a definite amount of nitrogen and phosphate which was to be used. Similarly, in our variety and in our cultivation tests we have quite arbitrarily set the level of fertilization.



It is not unlikely that the response to varied amounts of one nutrient may be different when tested at different levels of another nutrient. Thus, conclusions on the optimum amount may not be safely applied in a field practice that differed in its amounts of the other nutrients from those under which such optimum was determined. And it is quite possible that variety superiority may be indicated at one level of fertilization and may not be noted at some other level.

Thus there is a growing interest in the factorial type of field experiment in which two or more factors are compared in all possible combinations of their several different levels. Results are thus obtained on the response from the different factors singly, and at the same time upon their respective interactions, i.e., the way in which a change in one factor is influenced by a change in another.

Even though such interactions do not actually occur, and the response to one factor is substantially the same for all variations of another factor, the factorial experiment makes it possible to use a greater number of plots in making estimates of the effect of the different factors, and this greatly increases the precision with which the results can be measured. For instance, with 3 amounts of nitrogen and 3 amounts of potash, there are 9 possible treatment-combinations. If we identify the 3 nitrogen levels as N1, N2, and N3, and the 3 potash levels as K1, K2, and K3, these 9 treatment combinations will be:

N1 K1	N1 K2	N1 K3
N2 K1	N2 K2	N2 K3
N3 K1	N3 K2	N3 K3

With 6 replications for these 9 combinations, from an area with 54 plots, we could have a mean yield for each nitrogen level as secured from 18 plots, and also a mean yield for each potash level from 18 plots. This is possible because the potash differentials occur an equal number of times with each nitrogen level, and similarly, the nitrogen differentials occur an equal number of times with each potash level. If two separate tests had been installed on these 54 plots (27 plots for each test) then our mean yields would have come from only 9 plots for each level of N and of K. Thus this factorial plan would give us doubled precision for determining the effect of the 3 levels of both N and of K.

This comparative precision in estimating the effects of treatments in separate vs. factorial experiments may also be illustrated as follows: Our minimum "Grade A" standards call for testing (a) varieties with 5 replicates, and (b) fertilizer treatments with 7 replicates. Hence to test 2 varieties and 2 fertilizer treatments in separate experiments we need at least 24 plots. If we use these same 24 plots for a factorial experiment, combining both varieties with both fertilizers, we can have 12 plot yields to average for the effect of *each factor* (variety and fertilizer), and the estimate of the experimental error will be based on more than double the number of degrees of freedom obtainable in either single test.

Without further discussion of the principles involved in factorial designs, we give a few simple examples, using the analysis of variance therewith:

## EXAMPLE No. 1

A factorial experiment for 2 factors (X and Y) each at 2 levels (1 and 2); therefore, 4 combined Treatments in a balanced arrangement in 7 Blocks carrying a total of 28 plots.

Plot identity	Treatment combination		Assumed factors and amounts
A .....	X1	Y1	(125 lb N, 100 lb K <sub>2</sub> O)
B .....	X1	Y2	(125 lb N, 200 lb K <sub>2</sub> O)
C .....	X2	Y1	(175 lb N, 100 lb K <sub>2</sub> O)
D .....	X2	Y2	(175 lb N, 200 lb K <sub>2</sub> O)

## PLOT ARRANGEMENT AND YIELDS (T.C.A.)

I	A 70	C 79
	B 74	D 79
II	D 86	B 81
	C 74	A 70
III	C 80	A 78
	D 82	B 75
IV	B 85	D 88
	A 88	C 80
V	A 79	C 74
	B 76	D 80
VI	D 83	B 76
	C 71	A 79
VII	C 72	A 84
	D 79	B 72

## SET-UP FOR TOTALS

Block No.	A	B	C	D	Block totals
I .....	70	74	79	79	302
II .....	70	81	74	86	311
III .....	78	75	80	82	315
IV .....	88	85	80	88	341
V .....	79	76	74	80	309
VI .....	79	76	71	83	309
VII .....	84	72	72	79	307

Treatment totals 548 539 530 577 2194

Nitrogen totals:

$$125 \text{ lb (A + B)} = 548 + 539 = 1087$$

$$175 \text{ lb (C + D)} = 530 + 577 = 1107$$

Potash totals:

$$100 \text{ lb (A + C)} = 548 + 530 = 1078$$

$$200 \text{ lb (B + D)} = 539 + 577 = 1116$$

## Work Sheet for Example No. 1:

$$\text{Correction factor: } \frac{(2194)^2}{28} = 171916$$

Total Sum-Squares:

$$(70)^2 + (70)^2 + (78)^2 + (88)^2 + \dots (83)^2 + (79)^2 = \frac{172642}{-171916}$$

Corrected 726 (with 27 degrees of freedom)

Sum-Squares for Blocks:

$$\frac{(302)^2 + (311)^2 + (315)^2 + \dots (307)^2}{4} = \frac{688642}{4} = \frac{172161}{-171916}$$

Corrected 245 (with 6 degrees of freedom)

*Sum-Squares for Treatments:*

$$\frac{(548)^2 + (539)^2 + (530)^2 + (577)^2}{7} = \frac{1204654}{7} = 172093$$

—171916

Corrected 177 (with 3 degrees of freedom)

*Sum-Squares for Nitrogen:*

$$\frac{(1087)^2 + (1107)^2}{14} = \frac{2407018}{14} = 171930$$

—171916

Corrected 14 (with 1 degree of freedom)

*Sum-Squares for Potash:*

$$\frac{(1078)^2 + (1116)^2}{14} = \frac{2407540}{14} = 171967$$

—171916

Corrected 51 (with 1 degree of freedom)

*Sum-Squares for Interaction (between N and K):*

$$177 - (14 + 51) = 112 \text{ (with 1 degree of freedom)}$$

*Sum-Squares for Error:*

$$726 - (245 + 14 + 51 + 112) = 304 \text{ (with 18 degrees of freedom)}$$

From these data, we then set up the pertinent items in a table for the analysis of the variance as follows:

Source of variation	Degrees of freedom	Sum squares	Mean square or variance	"F"		Remarks
				Found	Required	
Blocks .....	6	245	.....	.....	.....	.....
Nitrogen .....	1	14	14.0	.83	4.41	Not significant
Potash .....	1	51	51.0	3.02	4.41	Not significant
Interaction (Nitrogen and Potash) .....	1	112	112.0	6.63	4.41	Significant
Error .....	18	304	16.9	.....	.....	.....
Total.....	27	726				

The "F" required to indicate a significant effect for either factor or their interaction is 4.41. Since the "F" value as found for nitrogen is only  $\frac{14.0}{16.9}$  or .83, and that for potash only  $\frac{51.0}{16.9}$  or 3.02, it would appear that the separate effects of nitrogen and potash are not significant. But there is a definite effect from their interaction  $\frac{112.0}{16.9} = 6.63$ , so the Treatment totals will need further study.

A difference between the combined treatment totals of

$t\sqrt{\text{Error Mean Square} \times n \times 2}$  or  $2.10\sqrt{16.9 \times 7 \times 2}$ , which amounts to 32.3 tons would be a significant amount. Hence we note these comparisons between the combined treatment totals:

"A" over "B":  $548 - 539 = 9$  tons Not significant

"A" over "C":  $548 - 530 = 18$  tons Not significant

"B" over "C":  $539 - 530 = 9$  tons Not significant

"D" over "A":  $577 - 548 = 29$  tons Not significant

"D" over "B":  $577 - 539 = 38$  tons (Avg. 5.43 T.C.A.) Significant

"D" over "C":  $577 - 530 = 47$  tons (Avg. 6.71 T.C.A.) Significant

Our interpretation of the results then becomes: (a) That there is a definite yield increase for 175 lbs. of nitrogen over 125 lbs. when potash is supplied at 200 lbs. ("D" over "B"), but not when potash is at only 100 lbs. ("C" over "A"); and (b) that there is also a gain for 200 lbs. of  $K_2O$  over 100 lbs. when nitrogen is supplied at 175 lbs. ("D" over "C") but not when only 125 lbs. of N is given ("B" over "A").

Thus it is apparent that the factorial plan has made it possible to show real effects from nitrogen and potash applications which might have been lost had the issues been tested in separate experiments.

### EXAMPLE NO. 2

Our second example is another  $2 \times 2$  Factorial Experiment for 2 Factors each at 2 levels; therefore, with 4 combined treatments arranged on 32 plots in 2 Latin Squares:

2 Factors = P and K  
2 Levels = 0 and 200 lbs.

Therefore 4 combinations, identified as.....

Plot identity	Amounts—	
	lb $P_2O_5$	lb $K_2O$
N =	0	0
NP =	200	0
NK =	0	200
NPK =	200	200

Plot numbers and identities, with yields as T.C.A.

Square I

Square II

1 NK 74	2 NPK 80	9 N 79	10 NP 71	17 NPK 78	18 NK 72	25 NP 77	26 N 71
3 N 69	4 NP 73	11 NK 78	12 NPK 84	19 NP 75	20 N 79	27 NPK 86	28 NK 76
5 NP 71	6 N 67	13 NPK 82	14 NK 74	21 N 73	22 NP 81	29 NK 84	30 NPK 82
7 NPK 76	8 NK 76	15 NP 69	16 N 77	23 NK 78	24 NPK 80	31 N 73	32 NP 75

The complete work sheet leading up to the analysis of variance is offered without further explanations:

#### Work Sheet for Example No. 2:

Square	Treatments				Position		
	NK	NPK	N	NP	Row totals	Column totals	Square totals
I	74	80	69	73	304	290	
	76	76	67	71	304	296	
	78	84	79	71	294	308	
	74	82	77	69	298	306	
Total .....	302	322	292	284	1200	1200	1200
II	72	78	79	75	298	304	
	78	80	73	81	316	312	
	76	86	71	77	320	320	
	84	82	73	75	306	304	
Total .....	310	326	296	308	1240	1240	1240
Treatment totals ..	612	648	588	592	Grand total .....		
Phosphate totals: "0" = 612 + 588 = 1200					Potash totals: "0" = 588 + 592 = 1180		
"200" = 648 + 592 = 1240					"200" = 612 + 648 = 1260		



*Sum-Squares for Squares:*

$$\frac{(1200)^2 + (1240)^2}{16} = \frac{2777600}{16} = 186100$$

$$\begin{array}{r} 186100 \\ -186050 \\ \hline \text{Corrected} \quad 50 \end{array}$$

*Sum-Squares for Treatments in Two Squares:*

$$\frac{(302)^2 + (310)^2 + (322)^2 + (326)^2 + (292)^2 + (296)^2 + (284)^2 + (308)^2}{4} =$$

$$\begin{array}{r} 745664 \\ 4 \quad = \quad 186416 \\ -186050 \\ \hline \text{Corrected} \quad 366 \end{array}$$

*Sum-Squares for Interaction (Treatments and Squares):*

$$366 - (50 + 282) = 34$$

*Sum-Squares for Error:*

$$694 - (282 + 92 + 98 + 50 + 34) = 694 - 556 = 138$$

#### ANALYSIS OF VARIANCE

Source	Degrees of freedom	Sum of squares	Mean square	"F"		Remarks
				Found	Required	
Rows . . . . .	(3+3) 6	92	...	....	....	....
Columns . . . . .	(3+3) 6	98	...	....	....	....
Squares . . . . .	1	50	...	....	....	....
Phosphate . . . . .	1	50	50	4.3	4.75	Not significant
Potash . . . . .	1	200	200	17.4	4.75	Significant
Interaction (Phosphate and Potash) . . . . .	1	32	32	2.8	4.75	Not significant
Interaction (Treatment and Squares) . . . . .	3	34	...	....	....	....
Error . . . . .	12	138	11.5	....	....	....
Total . . . . .	31	694				

Since the effect of potash is thus shown to be significant, a significant amount of difference between the potash totals would be  $t\sqrt{\text{Error Mean Square} \times n \times 2}$  or  $2.18\sqrt{11.5 \times 16 \times 2}$  or 41.8 tons.

Thus the difference (1260 — 1180) of 80 tons (an average for the 16 plots of 5 T.C.A.) is indicated to be a reliable gain that is most probably the result of the potash fertilization.

The response to phosphate is not significant, and there is no indication that a response from either one of these minerals is affected by the presence or absence of the other, i.e., there is no interaction between phosphate and potash.

#### EXAMPLE NO. 3

Example No. 3 is of a  $2 \times 3$  Factorial Experiment in which any 2 Factors\* ("A" and "B") are tested at all of any 3 Amounts ("1," "2," and "3"). Thus there are 6 combined Treatments, as follows:

A1	A2	A3
B1	B2	B3

\* Such as nitrogen, or phosphate, or potash, or lime, or forms, or methods, or varieties, etc.

These 6 treatments are shown in a balanced arrangement in 5 Blocks with a total of 30 plots:

TREATMENT IDENTITY AND YIELDS AS T.S.A.					
Block I		Block II		Block III	
A1	7.1	B3	8.0	A1	7.3
B2	8.4	A2	7.4	B2	7.7
A3	8.7	B1	7.2	A3	9.6
B3	8.1	A1	6.9	B3	8.8
A2	8.0	B2	7.6	A2	7.7
B1	7.6	A3	8.4	B1	7.5
Block IV		Block V			
B3	8.0	A1	6.0		
A2	7.4	B2	7.5		
B1	7.2	A3	8.4		
A1	7.0	B3	8.4		
B2	7.8	A2	7.2		
A3	9.2	B1	7.4		

These data may be set up for Totals as follows:

Block	A1	A2	A3	B1	B2	B3	Block totals
I	7.1	8.0	8.7	7.6	8.4	8.1	47.9
II	6.9	7.4	8.4	7.2	7.6	8.0	45.5
III	7.3	7.7	9.6	7.5	7.7	8.8	48.6
IV	7.0	7.4	9.2	7.2	7.8	8.0	46.6
V	6.0	7.2	8.4	7.4	7.5	8.4	44.9
Treatment totals	34.3	37.7	44.3	36.9	39.0	41.3	233.5

$$\text{Factor "A" Totals} = 34.3 + 37.7 + 44.3 = 116.3$$

$$\text{Factor "B" Totals} = 36.9 + 39.0 + 41.3 = 117.2$$

$$\text{Amount "1" Totals} = 34.3 + 36.9 = 71.2$$

$$\text{Amount "2" Totals} = 37.7 + 39.0 = 76.7$$

$$\text{Amount "3" Totals} = 44.3 + 41.3 = 85.6$$

#### Work Sheet for Example No. 3:

$$\text{Correction factor: } \frac{(233.5)^2}{30} = 1817.41$$

#### Total Sum-Squares:

$$(7.1)^2 + (6.9)^2 + (7.3)^2 + \dots + (8.0)^2 + (8.4)^2 = 1833.33$$

$$-1817.41$$

$$\text{Corrected } 15.92 \text{ (with 29 degrees of freedom)}$$

#### Sum-Squares Blocks:

$$\frac{(47.9)^2 + (45.5)^2 + (48.6)^2 + (46.6)^2 + (44.9)^2}{6} = \frac{10914.19}{6} = 1819.03$$

$$-1817.41$$

$$\text{Corrected } 1.62 \text{ (with 4 degrees of freedom)}$$

#### Sum-Squares Treatments:

$$\frac{(34.3)^2 + (37.7)^2 + (44.3)^2 + (36.9)^2 + (39.0)^2 + (41.3)^2}{5} =$$

$$\frac{9148.57}{5} = 1829.71$$

$$-1817.41$$

$$\text{Corrected } 12.30 \text{ (with 5 degrees of freedom)}$$

*Sum-Squares Factors (A and B):*

$$\frac{(116.3)^2 + (117.2)^2}{15} = \frac{27261.53}{15} = 1817.44$$

$$\frac{1817.44}{15} = 1817.41$$

Corrected .03 (with 1 degree of freedom)

*Sum-Squares Amounts (1, 2, and 3):*

$$\frac{(71.2)^2 + (76.7)^2 + (85.6)^2}{10} = \frac{18279.69}{10} = 1827.97$$

$$\frac{1827.97}{10} = 1817.41$$

Corrected 10.56 (with 2 degrees of freedom)

*Sum-Squares Interaction (Factors and Amounts):*

$$12.30 - (.03 + 10.56) = 1.71 \text{ [with } 5 - (1 + 2) \text{ or 2 degrees of freedom]}$$

*Sum-Squares Error:*

$$15.92 - (1.62 + .03 + 10.56 + 1.71) = 2.00$$

[with 29 - (4 + 1 + 2 + 2) or 20 degrees of freedom]

#### ANALYSIS OF VARIANCE

Source of variation	Degrees of freedom	Sum squares	Mean square or variance	"F"		Remarks
				Found	Required	
Blocks . . . . .	4	1.62	. . . .	. . . .	. . . .	. . . .
"Factors" . . . .	1	.03	.03	.3	4.35	Not significant
"Amounts" . . . .	2	10.56	5.28	52.8	3.49	Significant
Interaction . . . .	2	1.71	.86	8.6	3.49	Significant
Error . . . . .	20	2.00	.10	. . . .	. . . .	. . . .
Total . . . . .	29	15.92				

This analysis indicates that the "Factors" themselves do not show a significant difference, but that the "Amounts" do have real different effects upon the yields. A difference between the "Amounts" totals greater than

$t\sqrt{\text{Error Mean Square} \times n \times 2}$  or  $2.09\sqrt{.10 \times 10 \times 2}$  which is 2.95 tons would be significant. Hence it is clear that both "2" and "3" are better than "1," and also that "3" is better than "2."

Since this analysis also shows a definite interaction between the two factors and the amounts at which they were tested, we may look immediately for this interaction. A significant amount of difference between any two of the combined-treat-

ment totals would be  $t\sqrt{\text{Error Mean Square} \times n \times 2}$  or  $2.09\sqrt{.10 \times 5 \times 2}$  which amounts to 2.09 tons. A comparison of the various combined-treatment totals shows which differences are significant. Thus, Factor "B" is superior to "A" by  $(36.9 - 34.3)$  2.6 or an average of  $(2.6 \div 5)$  .52 T.S.A. when the Amount is "1," but when the Amount is "3" we note just the reverse situation, i.e., "A" is better than "B" by an average of  $\left(\frac{44.3 - 41.3}{5}\right)$  .60 T.S.A. However, with the Amount at "2" the two factors do not differ significantly.

#### EXAMPLE No. 4

For our fourth example, we use a  $4 \times 2$  Factorial Experiment which would be suitable for testing 4 Methods (A, B, C, D) of using 2 Amounts of Nitrogen (1, 2).



The 8 possible treatment combinations, i.e., A1, B1, C1, D1, A2, B2, C2, D2, might be laid down in an arrangement in 4 Blocks with 32 plots as follows:

YIELDS AS T.S.A.				IDENTITIES			
I	A1 5.1	D2 5.2	C2 6.0	B1 6.0	A = applied in 1 dose, at 3½ mos. B = applied in 2 doses, at 1½ and 3½ mos. C = applied in 3 doses, at 1½, 3½, and 6 mos. D = applied in 3 doses, at 1½, 3½, and 10 mos. 1 = 150 lb N 2 = 200 lb N		
	B2 5.8	C1 6.4	D1 5.6	A2 4.8			
II	C2 6.1	B1 5.7	A1 5.5	D2 5.4			
	D1 5.1	A2 5.7	B2 5.9	C1 6.9			
III	A1 4.9	D2 5.7	C2 5.6	B1 5.9			
	B2 6.1	C1 6.0	D1 5.9	A2 5.3			
IV	C2 5.2	B1 5.1	A1 4.6	D2 4.9			
	D1 5.0	A2 4.8	B2 5.4	C1 5.9			

#### ARRANGEMENT OF YIELD DATA TO SECURE TOTALS

Block	A1	B1	C1	D1	A2	B2	C2	D2	Block totals
I .....	5.1	6.0	6.4	5.6	4.8	5.8	6.0	5.2	44.9
II .....	5.5	5.7	6.9	5.1	5.7	5.9	6.1	5.4	46.3
III .....	4.9	5.9	6.0	5.9	5.3	6.1	5.6	5.7	45.4
IV .....	4.6	5.1	5.9	5.0	4.8	5.4	5.2	4.9	40.9
Treatment totals ...	20.1	22.7	25.2	21.6	20.6	23.2	22.9	21.2	177.5

“Methods” totals:

$$“A” = 20.1 + 20.6 = 40.7$$

$$“B” = 22.7 + 23.2 = 45.9$$

$$“C” = 25.2 + 22.9 = 48.1$$

$$“D” = 21.6 + 21.2 = 42.8$$

“Nitrogen” totals:

$$“1” = 20.1 + 22.7 + 25.2 + 21.6 = 89.6$$

$$“2” = 20.6 + 23.2 + 22.9 + 21.2 = 87.9$$

#### Work Sheet for Example No. 4:

$$\text{Correction factor: } \frac{(177.5)^2}{32} = 984.57$$

Total Sum-Squares:

$$(5.1)^2 + (5.5)^2 + (4.9)^2 + \dots (5.7)^2 + (4.9)^2 = 993.09$$

$$-984.57$$

Corrected 8.52 (with 31 degrees of freedom)

Sum-Squares Blocks:

$$\frac{(44.9)^2 + (46.3)^2 + (45.4)^2 + (40.9)^2}{8} = \frac{7893.67}{8} = 986.71$$

$$-984.57$$

Corrected 2.14 (with 3 degrees of freedom)

Sum-Squares Treatments:

$$\frac{(20.1)^2 + (22.7)^2 + (25.2)^2 + \dots (21.2)^2}{4} = \frac{3957.35}{4} = 989.34$$

$$-984.57$$

Corrected 4.77 (with 7 degrees of freedom)



## YIELDS AS T.S.A.

G. L. SQUARE I

G. L. SQUARE II

A1 8.01	B2 8.60	X3 7.65	B2 9.99	A1 8.22	X3 8.71
B3 8.26	X1 7.41	A2 8.67	X1 8.00	B3 9.27	A2 9.61
X2 7.62	A3 8.83	B1 8.04	A3 9.14	X2 8.61	B1 8.18
B2 9.64	X3 9.02	A1 8.13	G. L. S. III		
X1 8.43	A2 9.06	B3 9.10			
A3 8.72	B1 8.77	X2 8.60			
X3 8.37	A1 8.10	B2 9.01	G. L. S. IV		
A2 8.76	B3 8.34	X1 7.64			
B1 7.83	X2 8.48	A3 8.61			

These data may be set up as follows :

Treatments	Square I	Square II	Square III	Square IV	Treatment totals	Variety totals
A1 .....	8.01	8.22	8.13	8.10	32.46	
A2 .....	8.67	9.61	9.06	8.76	36.10	
A3 .....	8.83	9.14	8.72	8.61	35.30	
Total A .....						103.86
B1 .....	8.04	8.18	8.77	7.83	32.82	
B2 .....	8.60	9.99	9.64	9.01	37.24	
B3 .....	8.26	9.27	9.10	8.34	34.97	
Total B .....						105.03
X1 .....	7.41	8.00	8.43	7.64	31.48	
X2 .....	7.62	8.61	8.60	8.48	33.31	
X3 .....	7.65	8.71	9.02	8.37	33.75	
Total X .....						98.54
Square totals .....	73.09	79.73	79.47	75.14		
Grand total .....						307.43

## Nitrogen Totals

$$''1'' = (32.46 + 32.82 + 31.48) = 96.76$$

$$''2'' = (36.10 + 37.24 + 33.31) = 106.65$$

$$''3'' = (35.30 + 34.97 + 33.75) = 104.02$$

## Work Sheet for Example No. 5:

$$\text{Correction factor: } \frac{(307.43)^2}{36} = 2625.37$$

## Total Sum-Squares:

$$(8.01)^2 + (8.67)^2 + (8.83)^2 + (8.04)^2 + \dots (8.48)^2 + (8.37)^2 =$$

$$\frac{2637.84}{-2625.37}$$

Corrected 12.47 (with 35 degrees of freedom)

*Sum-Squares for Graeco-Latin Squares:*

$$\frac{(73.09)^2 + (79.73)^2 + (79.47)^2 + (75.14)^2}{9} = \frac{23660.52}{9} = 2628.95$$

$$\begin{array}{r} 2628.95 \\ -2625.37 \\ \hline \text{Corrected} \quad 3.58 \text{ (with 3 degrees of freedom)} \end{array}$$

*Sum-Squares Treatments:*

$$\frac{(32.46)^2 + (36.10)^2 + (35.30)^2 + \dots (33.75)^2}{4} = \frac{10529.43}{4} = 2632.36$$

$$\begin{array}{r} 2632.36 \\ -2625.37 \\ \hline \text{Corrected} \quad 6.99 \text{ (with 8 degrees of freedom)} \end{array}$$

*Sum-Squares Varieties only:*

$$\frac{(103.86)^2 + (105.03)^2 + (98.54)^2}{12} = \frac{31528.33}{12} = 2627.36$$

$$\begin{array}{r} 2627.36 \\ -2625.37 \\ \hline \text{Corrected} \quad 1.99 \text{ (with 2 degrees of freedom)} \end{array}$$

*Sum-Squares Nitrogen only:*

$$\frac{(96.76)^2 + (106.65)^2 + (104.02)^2}{12} = \frac{31556.88}{12} = 2629.74$$

$$\begin{array}{r} 2629.74 \\ -2625.37 \\ \hline \text{Corrected} \quad 4.37 \text{ (with 2 degrees of freedom)} \end{array}$$

*Sum-Squares for Interaction (Varieties and Nitrogen):*

$$6.99 - (1.99 + 4.37) = .63 \text{ [with } 8 - (2 + 2) \text{ or 4 degrees of freedom]}$$

*Sum-Squares for Error:*

$$12.47 - (3.58 + 1.99 + 4.37 + .63) = 1.90 \text{ [with } 35 - (3 + 2 + 2 + 4) \text{ or 24 degrees of freedom]}$$

## ANALYSIS OF VARIANCE

Source	Degrees of freedom	Sum of squares	Mean square	"F"		Remarks
				Found	Required	
Graeco-Latin Squares..	3	3.58	....	....	....	....
Varieties .....	2	1.99	1.00	12.5	3.40	Significant
Nitrogen .....	2	4.37	2.19	27.4	3.40	Significant
Interaction (V × N) ..	4	.63	.16	2.0	2.78	Not significant
Error .....	24	1.90	.08	....	....	....
Total.....	35	12.47				

There is no interaction between these 3 varieties and the amounts of nitrogen at which they were tested.

A significant amount of difference ( $t \times \text{SEd}$ ) for the Variety and Nitrogen totals would be  $2.06\sqrt{.08 \times 12 \times 2}$  or 2.86 tons.

Hence we find both varieties "A" and "B" better than "X" but not dissimilar between themselves; also nitrogen levels "2" and "3" better than "1," but no real difference between "2" and "3."

(This same arrangement is a proper one for the analysis of variance as made for multiple Latin squares. In such a case the detail would be set up with degrees of freedom as follows:

Source	Degrees of freedom	
Squares .....	3	
Rows (2 + 2 + 2 + 2) .....	8	
Columns (2 + 2 + 2 + 2) .....	8	
Treatments .....	8	$\left\{ \begin{array}{l} \text{Varieties } 2 \\ \text{Nitrogen } 2 \\ \text{Interaction (V} \times \text{N)} \ 4 \end{array} \right.$
Error .....	8	
Total.....	35	

Unless the mean variance for rows and columns is quite large, it may be best to leave same with the Error variance to insure a more conservative estimate of the treatment effects. Thus one can "go too far" in breaking down the Error variance, and so reduce this measure to a point beyond where it is practically no measure at all.)

#### EXAMPLE NO. 6

For our sixth example we have selected a plan for a  $4 \times 3$  Factorial experiment which should be suitable for testing 4 varieties (A, B, C, D) with 3 Amounts of Nitrogen (1, 2, 3). This calls for 12 combined Treatments:

A1	A2	A3
B1	B2	B3
C1	C2	C3
D1	D2	D3

These 12 treatments may best be installed in a split-plot arrangement. We have used 4 blocks, with 16 whole-plots divided so as to furnish 48 split-plots.

In any split-plot arrangement, those factors between which the greater yield differences are to be expected should be tested in the whole-plots, while those for which the smaller differences are expected will be tested in the split-plots. This is desirable since the number of replications of the split-plots is greater than the replicates of whole-plots:

Block 1				Block 2				Block 3				Block 4			
A1 66	B3 80	C3 80	D1 61	C1 62	B3 80	A3 81	D1 69	C1 62	B3 80	A3 81	D1 69	C1 62	B3 80	A3 81	D1 69
A2 77	B2 74	C2 81	D2 82	C2 88	B2 82	A2 76	D2 72	C2 88	B2 82	A2 76	D2 72	C2 88	B2 82	A2 76	D2 72
A3 79	B1 71	C1 70	D3 81	C3 81	B1 79	A1 63	D3 74	C3 81	B1 79	A1 63	D3 74	C3 81	B1 79	A1 63	D3 74
D3 77	C1 72	B1 83	A3 85	D3 84	A1 73	B1 75	C3 73	D3 84	A1 73	B1 75	C3 73	D3 84	A1 73	B1 75	C3 73
D2 77	C2 81	B2 78	A2 83	D2 77	A2 84	B2 78	C2 74	D2 77	A2 84	B2 78	C2 74	D2 77	A2 84	B2 78	C2 74
D1 60	C3 74	B3 75	A1 70	D1 70	A3 83	B3 85	C1 64	D1 70	A3 83	B3 85	C1 64	D1 70	A3 83	B3 85	C1 64

## DETAILS FOR THIS ANALYSIS OF VARIANCE

Source	Degrees of freedom	
Blocks . . . . .	3	
Varieties . . . . .	3	
Error (a) . . . . .	9	(Total degrees of freedom for whole plots = 15)
<hr/>		
Nitrogen . . . . .	2	
Interaction (3 × 2) (Varieties and Nitrogen)	6	
Error (b) . . . . .	24	
<hr/>		
Total . . . . .	47	

Note that:

- (1) Degrees of freedom for Error (a) = Degrees of freedom for Blocks × Degrees of Freedom for Varieties, e.g.,  $3 \times 3$ .
- (2) Degrees of freedom for Error (b) = Degrees of freedom for Blocks × Sum of Degrees of Freedom for both Nitrogen and Interaction, e.g.,  $3 \times (2 + 6)$ .

## SET-UP FOR SECURING TOTALS

Varieties	Block No.	—Nitrogen Treatment—			Whole plot totals	Variety totals
		No. 1	No. 2	No. 3		
A . . . . .	1	66	77	79	222	
	2	70	83	85	238	
	3	73	84	83	240	
	4	63	76	81	220	
Total . . . . .		272	320	328		920
B . . . . .	1	71	74	80	225	
	2	83	78	75	236	
	3	79	82	80	241	
	4	75	78	85	238	
Total . . . . .		308	312	320		940
C . . . . .	1	72	81	74	227	
	2	70	81	80	231	
	3	62	88	81	231	
	4	64	74	73	211	
Total . . . . .		268	324	308		900
D . . . . .	1	60	77	77	214	
	2	61	82	81	224	
	3	70	77	84	231	
	4	69	72	74	215	
Total . . . . .		260	308	316		884
Nitrogen totals . . .		1108	1264	1272	Grand total . .	3644
Blocks:		1	2	3	4	
		222	238	240	220	
		225	236	241	238	
		227	231	231	211	
		214	224	231	215	
Block totals . . . . .		888	929	943	884	(3644)

**Work Sheet for Example No. 6:**

$$\text{Correction factor: } \frac{(3644)^2}{48} = 276640$$

**Total Sum-Squares:**

$$(66)^2 + (70)^2 + (73)^2 + (63)^2 + (71)^2 + \dots (84)^2 + (74)^2 = \frac{278836}{-276640}$$

Corrected 2196 (with 47 degrees of freedom)

**Sum-Squares for Blocks:**

$$\frac{(888)^2 + (929)^2 + (943)^2 + (884)^2}{12} = 276858$$

$$-276640$$

Corrected 218 (with 3 degrees of freedom)

**Sum-Squares for Whole Plots:**

$$\frac{(222)^2 + (238)^2 + (240)^2 + \dots (231)^2 + (215)^2}{3} = 277101$$

$$-276640$$

Corrected 461 (with 15 degrees of freedom)

**Sum-Squares for Varieties:**

$$\frac{(920)^2 + (940)^2 + (900)^2 + (884)^2}{12} = 276788$$

$$-276640$$

Corrected 148 (with 3 degrees of freedom)

**Sum-Squares for Error (a) (for Varieties):**

$$461 - (218 + 148) = 95 \text{ [with } 15 - (3 + 3) \text{ or 9 degrees of freedom]}$$

**Sum-Squares for Treatments:**

$$\frac{(272)^2 + (308)^2 + (268)^2 + \dots (308)^2 + (316)^2}{4} = 278140$$

$$-276640$$

Corrected 1500 (with 11 degrees of freedom)

**Sum-Squares for Nitrogen:**

$$\frac{(1108)^2 + (1264)^2 + (1272)^2}{16} = 277709$$

$$-276640$$

Corrected 1069 (with 2 degrees of freedom)

**Sum-Squares for Interaction (V × N):**

$$1500 - (148 + 1069) = 283 \text{ [with } 11 - (3 + 2) \text{ or 6 degrees of freedom]}$$

**Sum-Squares Error (b) (for Nitrogen and Interaction):**

$$2196 - (218 + 148 + 95 + 1069 + 283) = 383$$

[with 47 - (3 + 3 + 9 + 2 + 6) or 24 degrees of freedom]

**ANALYSIS OF VARIANCE**

Source of variation	Degrees of freedom	Total sum squares	Mean square	"F"		Remarks
				Found	Required	
Blocks .....	3	218	....	....	....	....
Varieties .....	3	148	49.3	4.65	3.86	Significant
Error (a).....	9	95	10.6	....	....	....
Nitrogen .....	2	1069	534.5	33.41	3.40	Significant
Interaction (V × N) ..	6	283	47.2	2.95	2.51	Significant
Error (b) .....	24	383	16.0	....	....	....
Total.....	47	2196				

For a significant amount of difference between the 3 Variety totals we would need  $t\sqrt{\text{Error (a) Mean Square} \times n \times 2}$  or  $2.26\sqrt{10.6 \times 12 \times 2}$  which is 36 tons. Hence we note the following:

"B" is better than "C" or "D" but not better than "A."

"A" is also probably superior to "D" but not to "C."

There is no difference between "D" and "C."

A significant amount of difference between the 3 Nitrogen totals would be  $t\sqrt{\text{Error (b) Mean Square} \times n \times 2}$  or  $2.06\sqrt{16.0 \times 16 \times 2}$  which is 46.6 tons. Thus Nitrogen Nos. 2 and 3 are both better than No. 1, but Nos. 2 and 3 are not significantly different themselves.

The evidence of a significant interaction between the varieties and nitrogen suggests that their response is not always the same, and so we look further to see just what this difference is. A significant amount of difference between the 12 treatment totals would be  $2.06\sqrt{16.0 \times 4 \times 2}$  or 23.3 tons. And so we note that Variety "B" is superior to "A," "C," and "D" with Nitrogen No. 1 but not with the other two amounts of nitrogen.

#### EXAMPLE No. 7

For the seventh example, we apply the analysis of variance to the results which have been secured from the same group of plots which have received the same treatment for a series of years: in other words, to the analysis of a Serial Experiment. This procedure should be an exceedingly valuable one for the plantation agriculturist who has accumulated results of this nature and has been somewhat at a loss to interpret them collectively.

For this example we have used an experiment in which there were 3 Treatments (R, S, T) which had been harvested for 4 years (31, 33, 35, 37). The data were secured from an arrangement of 21 plots in 7 Blocks, as follows:

Block I			Block II			Block III			Block IV		
T	S	R	T	S	R	T	S	R	T	S	R
			R	S	T	R	S	T	R	S	T
Block V			Block VI			Block VII					

The yields for each Treatment in each Block for each Year are arranged for totals as follows:

Year	Block	Treatment	Treatment	Treatment	Block
		R	S	T	totals
1931	1	85	86	93	264
	2	92	86	89	267
	3	86	94	89	269
	4	91	88	76	255
	5	89	86	94	269
	6	83	91	87	261
	7	80	88	79	247
Treatment totals . . . . .		606	619	607	1832 Year total



		Treatment	Treatment	Treatment	Block	
1933	1	85	84	101	270	
	2	86	99	93	278	
	3	85	94	104	283	
	4	88	85	99	272	
	5	83	97	91	271	
	6	83	89	98	270	
	7	82	99	84	265	
		<hr/>	<hr/>	<hr/>	<hr/>	
	Treatment totals .....	592	647	670	1909	Year total
1935	1	111	94	111	316	
	2	89	107	102	298	
	3	99	90	112	301	
	4	82	89	108	279	
	5	87	94	97	278	
	6	95	97	106	298	
	7	78	112	88	278	
		<hr/>	<hr/>	<hr/>	<hr/>	
	Treatment totals .....	641	683	724	2048	Year total
1937	1	90	79	101	270	
	2	82	92	82	256	
	3	104	87	88	279	
	4	80	78	86	244	
	5	75	91	88	254	
	6	73	83	99	255	
	7	74	96	75	245	
		<hr/>	<hr/>	<hr/>	<hr/>	
	Treatment totals .....	578	606	619	1803	Year total
		<hr/>	<hr/>	<hr/>	<hr/>	
	Treatment totals for four years .....	2417	2555	2620	7592	Grand total

**Work Sheet for Example No. 7:**

$$\text{Correction factor: } \frac{(7592)^2}{84} = \frac{57638464}{84} = 686172$$

**Total Sum-Squares:**

$$(85)^2 + (92)^2 + (86)^2 + (91)^2 + \dots (99)^2 + (75)^2 = 693142$$

$$\underline{-686172}$$

Corrected 6970 (83 degrees of freedom)

**Sum-Squares Blocks and Years:**

$$\frac{(264)^2 + (267)^2 + (269)^2 + \dots (245)^2}{3} = \frac{2066538}{3} = 688846$$

$$\underline{-686172}$$

Corrected 2674 (27 degrees of freedom)

**Sum-Squares Combined Years  $\times$  Treatments:**

$$\frac{(606)^2 + (592)^2 + (641)^2 + \dots (619)^2}{7} = \frac{4822846}{7} = 688978$$

$$\underline{-686172}$$

Corrected 2806 (11 degrees of freedom)

**Sum-Squares Treatments:**

$$\frac{(2417)^2 + (2555)^2 + (2620)^2}{28} = \frac{19234314}{28} = 686940$$

$$\underline{-686172}$$

Corrected 768 (2 degrees of freedom)

*Sum-Squares Years:*

$$\frac{(1832)^2 + (1909)^2 + (2048)^2 + (1803)^2}{21} = \frac{14445618}{21} = 687887$$

$$\frac{687887}{-686172}$$

$$\text{Corrected} \quad 1715 \text{ (3 degrees of freedom)}$$

*Sum-Squares Blocks:*

$$(\text{Sum-Squares Blocks and Years}) - (\text{Sum-Squares Years}) = 2674 - 1715 = 959$$

$$(27 - 3 = 24 \text{ degrees of freedom})$$

*Sum-Squares Interaction (Years  $\times$  Treatment):*

$$(\text{Sum-Squares Combined Years} \times \text{Treatments}) - (\text{Sum-Squares Years} + \text{Sum-Squares Treatments}) = 2806 - (1715 + 768) = 2806 - 2483 = 323$$

$$[11 - (3 + 2) = 6 \text{ degrees of freedom}]$$

*Sum-Squares Error:*

$$(\text{Total Sum-Squares}) - [\text{Sum-Squares Blocks} + \text{Sum-Squares Treatments} + \text{Sum-Squares Years} + \text{Sum-Squares Interaction (Y + T)}] = 6970 - (959 + 768 + 1715 + 323) = 6970 - 3765 = 3205$$

$$[83 - (24 + 2 + 3 + 6) = 48 \text{ degrees of freedom}]$$

## ANALYSIS OF VARIANCE

Source	Degrees of freedom	Sum squares	Mean square	"F"		Remarks
				Found	Required	
Blocks .....	24	959	....	....	....	....
Treatments .....	2	768	384	5.73	3.18	Significant
Years .....	3	1715	572	8.54	2.79	Significant
Interaction (treatments $\times$ years) .....	6	323	54	.81	2.29	No effect
Error .....	48	3205	67	....	....	....
Total .....	83	6970				

$$\text{SEd for Treatment Totals} = \sqrt{\text{Error Mean Square} \times n \times 2}$$

$$= \sqrt{67 \times 28 \times 2} = \sqrt{3752} = 61.3$$

$$\text{Amount needed for significance} = t \times \text{SEd} = 2.01 \times 61.3 = 123.2 \text{ tons}$$

*Differences:*

$$\text{Treatment S over R} = 2555 - 2417 = 138 \text{ tons (Avg. } \frac{138}{28} = 4.93 \text{ T.C.A.) Significant}$$

$$\text{Treatment T over S} = 2620 - 2555 = 65 \text{ tons (Avg. } \frac{65}{28} = 2.32 \text{ T.C.A.) Not significant}$$

$$\text{SEd for Year Totals} = \sqrt{67 \times 21 \times 2} = \sqrt{2814} = 53.1$$

$$\text{For significance: } t \times \text{SEd} = 2.01 \times 53.1 = 106.7 \text{ tons}$$

Also, but of lesser interest, is the fact that the 1935 yields were definitely higher than those for 1933, 1931, and 1937; but there was no real difference between the 1931, 1933 and 1937 yields.

There was no effect of interaction between treatments and years.

## EXAMPLE NO. 8

For our eighth and final example, we offer a more complex Factorial Arrangement in which 3 Factors (N, P, K) are all combined at each of 2 Levels (1, 2):

this gives 8 combined Treatments which we have arranged on an area of 32 plots in 4 Blocks as indicated:

### YIELDS AS T.C.A.

Block I		Block II	
A 71	C 75	D 84	B 71
J 74	L 81	M 87	K 76
K 77	M 78	L 84	J 79
B 75	D 77	C 77	A 74
Block III			
C 72	A 70		
L 82	J 78		
M 80	K 71		
D 76	B 68		
Block IV			
B 70	D 64		
K 73	M 84		
J 72	L 79		
A 66	C 71		

Treatment  
identity

Combination

A .....	N1 + P1 + K1
B .....	N1 + P2 + K1
C .....	N1 + P1 + K2
D .....	N1 + P2 + K2
J .....	N2 + P1 + K1
K .....	N2 + P2 + K1
L .....	N2 + P1 + K2
M .....	N2 + P2 + K2

### PLOT YIELDS AS SET UP FOR TOTALS

Block	A	B	C	D	J	K	L	M	Block totals
I .....	71	75	75	77	74	77	81	78	608
II .....	74	71	77	84	79	76	84	87	632
III .....	70	68	72	76	78	71	82	80	597
IV .....	66	70	71	64	72	73	79	84	579
Treatment totals.....	281	284	295	301	303	297	326	329	2416

The next step is to secure the proper data for use in calculating the treatment effects for the main factors as well as for the 2- and 3-Factor combinations. This is most easily done by setting up the following tabular arrangement:

### COMBINATION OF TREATMENTS TO SHOW MAIN EFFECTS AND DOUBLE AND TRIPLE INTERACTIONS

Effect of	A	B	C	D	J	K	L	M
N .....	—	—	—	—	+	+	+	+
P .....	—	+	—	+	—	+	—	+
K .....	—	—	+	+	—	—	+	+
NP .....	+	—	+	—	—	+	—	+
NK .....	+	+	—	—	—	—	+	+
PK .....	+	—	—	+	+	—	—	+
NPK .....	—	+	+	—	+	—	—	+

This table must be correctly made up. The first step is to assign the proper + and — signs for each of the 3 main effects (N, P, and K): a — sign is placed in the proper columns under the Treatment Identity headings, for each Level "1,"

and a + sign for each Level "2" which is associated with each main effect factor indicated in the left-hand column.

The next step is to determine the + and — signs for the 2-factor effects (NP, NK, and PK). This is done (1) by assigning a + sign when the main effect signs of the two factors concerned are the same, i.e., when the main effect signs are both + or both —, and (2) by assigning a — sign when they are different.

Similarly, for the 3-factor effect (NPK), the sign will be + when its 2-factor and main effect both have the same sign, and — when they have different signs.

These signs are necessary in order to calculate the treatment effects shown in the next table. This table is made up by combining the treatment totals according to the + and — signs in the above plan for determining the main and interaction effects.

#### CALCULATION OF TREATMENT EFFECTS

For	A	B	C	D	J	K	L	M	Treatment effect*
N .....	—281	—284	—295	—301	+303	+297	+326	+329 =	94
P .....	—281	+284	—295	+301	—303	+297	—326	+329 =	6
K .....	—281	—284	+295	+301	—303	—297	+326	+329 =	86
NP .....	+281	—284	+295	—301	—303	+297	—326	+329 =	—12
NK .....	+281	+284	—295	—301	—303	—297	+326	+329 =	24
PK .....	+281	—284	—295	+301	+303	—297	—326	+329 =	12
NPK .....	—281	+284	+295	—301	+303	—297	—326	+329 =	6

\* The total difference between the higher and the lower level.

Perhaps these interaction effects will be more easily understood if we examine them from the properly chosen levels of the Treatment totals as follows:

Treatment identity	(Levels of:)			Total yield
	N	P	K	
A	1	1	1	281
B	1	2	1	284
C	1	1	2	295
D	1	2	2	301
J	2	1	1	303
K	2	2	1	297
L	2	1	2	326
M	2	2	2	329

(1) The total NP interaction will be the difference between the sum of those treatments which have a similar level of N and P, and the sum of the treatments having different levels of N and P, e.g.

$$(A + C + K + M) - (B + D + J + L) = 1202 - 1214 = -12 \text{ tons}$$

Furthermore, with P1, we have a gain for N2 over N1 of 53 tons:

$$(J + L) - (A + C) = 629 - 576 = 53,$$

while with P2, we have a gain for N2 over N1 of only 41 tons:

$$(K + M) - (B + D) = 626 - 585 = 41.$$

Hence the difference of N2 over N1 was less by  $(53 - 41)$  12 tons in the presence of high phosphate (P2) than with low phosphate (P1).

Conversely, with N1, we have a gain for P2 over P1 of 9 tons:

$$(B + D) - (A + C) = 585 - 576 = 9,$$

while with N2, we have a loss for P2 over P1 of  $-3$  tons:

$$(K + M) - (J + L) = 626 - 629 = -3.$$

Thus in the presence of high nitrogen (N2) there were 12 tons less cane from the high-phosphate (P2) than from the low-phosphate (P1) treatment.

(2) The total NK interaction will be the difference between the sum of the treatments having the same levels of N and K, and the sum of the treatments having different levels of N and K, e.g.

$$(A + B + L + M) - (C + D + J + K) = 1220 - 1196 = 24.$$

With K1, we have a gain of 35 tons for N2 over N1, e.g.

$$(J + K) - (A + B) = 600 - 565 = 35.$$

With K2, the gain for N2 over N1 was 59 tons, e.g.

$$(L + M) - (C + D) = 655 - 596 = 59.$$

Hence the difference of N2 over N1 was greater by  $(59 - 35)$  24 tons in the presence of high potash (K2) than with low potash (K1).

Similarly it may be shown that the difference between K2 and K1 was greater by 24 tons in the presence of high nitrogen (N2) than with low nitrogen (N1), e.g.

$$\begin{aligned}(C + D) - (A + B) &= 596 - 565 = 31 \\(L + M) - (J + K) &= 655 - 600 = 55 \\55 - 31 &= 24.\end{aligned}$$

(3) The total PK interaction amounting to 12 tons may be shown as follows:

$$(A + J + D + M) - (B + K + C + L) = 1214 - 1202 = 12.$$

With P1, there is a gain of 37 tons for K2 over K1, while with P2 this gain for K2 over K1 is 49 tons; hence a difference of 12 tons favors K2 over K1 with the higher phosphate. Similarly, the difference of  $-3$  tons for P2 over P1 with K1, as compared with a corresponding gain of 9 tons with K2, indicates a 12-ton difference favoring P2 over P1 in the presence of the higher potash level.

(4) The 3-factor interaction of N, P, and K can be shown to be the difference between (a) 2 levels of one factor when the level of the other two factors is similar, and (b) the same 2 levels of the same factor when the level of the other two factors is different, e.g.

(a) The difference between N2 and N1, when each is associated with the same level of both P and K, was 50 tons:

$$(M + J) - (A + D) = 632 - 582 = 50.$$

(b) The difference between N2 and N1, when associated with a different level of both P and K, was 44 tons:

$$(K + L) - (C + B) = 623 - 579 = 44.$$

The total NPK interaction is therefore the difference between these two amounts, 50 — 44 or 6 tons.

**Work Sheet for Example No. 8:**

$$\text{Correction factor: } \frac{(2416)^2}{32} = \frac{5837056}{32} = 182408$$

*Total Sum-Squares (with 31 degrees of freedom):*

$$(71)^2 + (74)^2 + (70)^2 + (66)^2 + (75)^2 + \dots (80)^2 + (84)^2 = \frac{183350}{-182408}$$

Corrected 942

*Sum-Squares Blocks (with 3 degrees of freedom):*

$$\frac{(608)^2 + (632)^2 + (597)^2 + (579)^2}{8} = \frac{1460738}{8} = 182592$$

—182408

Corrected 184

*Sum-Squares Treatments (with 7 degrees of freedom):*

$$\frac{(281)^2 + (284)^2 + (295)^2 + \dots (329)^2}{4} = \frac{731778}{4} = 182945$$

—182408

Corrected 537

*Sum-Squares Error (with 21 degrees of freedom):*

$$942 - (184 + 537) = 221$$

**PARTITION OF SUM SQUARES TREATMENTS\***

		Sum squares				
N	.....	$(94)^2 \div 32 =$	276.12	Note: The sum of these Sum Squares (537) must check with the Sum - Squares Treatments as previously calculated.		
P	.....	$(6)^2 \div 32 =$	1.13			
K	.....	$(86)^2 \div 32 =$	231.13			
NP	.....	$(12)^2 \div 32 =$	4.50			
NK	.....	$(24)^2 \div 32 =$	18.00			
PK	.....	$(12)^2 \div 32 =$	4.50			
NPK	.....	$(6)^2 \div 32 =$	1.13			
			536.51			

The Analysis of Variance may then be set up as follows:

Source	Degrees of freedom	Sum squares	Mean square	"F"		Remarks
				Found	Required	
Blocks	3	184	.....	.....	.....	.....
N only	1	276	276	26.3	4.32	Significant
P only	1	1	1	.1	4.32	Not significant
K only	1	231	231	22.0	4.32	Significant
N and P	1	5	5	.5	4.32	Not significant
N and K	1	18	18	1.7	4.32	Not significant
P and K	1	5	5	.5	4.32	Not significant
N, P, and K	1	1	1	.1	4.32	Not significant
Error	21	221	10.5	.....	.....	.....
Total	31	942				

\* (Treatment effect)<sup>2</sup> ÷ total number of plots.

Thus we note a reliable effect of nitrogen and of potash, but no significant interaction between them, nor is there any real influence shown by the phosphate applications.

The SEd for the main effect totals would be  $\sqrt{10.5 \times 16 \times 2}$  or 18.3 tons and a significant amount of difference between same would be  $2.08 \times 18.3$  or 38.1 tons. This makes both the difference between the two nitrogen levels (94) and that between the two potash levels (86) significant amounts; an average gain for the second levels over the first levels of  $\left(\frac{94}{16}\right)$  5.9 T.C.A. for nitrogen, and of  $\left(\frac{86}{16}\right)$  5.4 T.C.A. for potash.

Neither the 2-Factor effects nor the 3-Factor effect are significant. The total gain of 24 tons for the interaction of N and K is not a reliable one. (If any 2-Factor effect had been significant, its SEd would have been  $\sqrt{10.5 \times 8 \times 2}$  or 13 tons and a significant amount of difference between the 2-Factor effect totals would have been  $2.08 \times 13$  or 27 tons. And for a significant 3-Factor effect, a significant difference would have needed to be  $2.08\sqrt{10.5 \times 4 \times 2}$  or 19.1 tons.)

### DISCUSSION

"Student's" method for analyzing the results of field experiments has been very popular with our plantation agriculturists. That it has a common basis with Fisher's analysis of variance can be shown by applying it to the same set of data we have used on page 79, and determining the standard error of the mean difference between the paired plot yields. This error will be found to be the same as the standard error of the difference determined from the mean Error Variance of the Analysis of Variance calculation, by the formula

$$\text{SEd} = \sqrt{\frac{\text{Error Variance} \times 2}{n}}$$

For example (Student's Method):

		Difference			
	"A"	"X"	"A" over "X"	d	d <sup>2</sup>
	76	74	+ 2	.62	.3844
	81	79	+ 2	.62	.3844
	73	73	0	-1.38	1.9044
	76	71	+ 5	3.62	13.1044
	72	70	+ 2	.62	.3844
	77	83	- 6	-7.38	54.4644
	82	80	+ 2	.62	.3844
	74	70	+ 4	2.62	6.8644
Totals . . . . .	611	600	+11		77.8752
Average . . . . .	76.4	75.0			
Mean difference . . . . .			+ 1.38		

From these data we may calculate a standard error (SEm) for this mean difference (1.37) as follows:

$$SEm = \sqrt{\frac{\text{Sum of } d^2}{n(n-1)}} = \sqrt{\frac{77.8752}{8 \times 7}} = \sqrt{1.39} = 1.18$$

Similarly, the standard error of a difference (SEd) between the two means, from the analysis of variance calculation, would be found from the formula:

$$SEd = \sqrt{\frac{\text{Error Variance} \times 2}{n}} = \sqrt{\frac{5.57 \times 2}{8}} = \sqrt{1.39} = 1.18$$

Thus we note that both methods give the identical standard error for the difference between treatments. This fact holds good, however, only when 2 treatments are being compared. When more than 2 treatments are being tested in the same experiment, this similarity does not hold because the analysis of variance uses the generalized error as secured from the pooled sum of squares in place of the individual error from pairs of plots as obtained in Student's method.

There is still some difference of opinion with regard to the preference for the generalized error (as obtained from the analysis of variance) and the individual error (as secured from Student's method). The variance method assumes that a more or less homogeneous error is associated with all treatments, and we know that this assumption may not always be true for differential treatments included in field tests with sugar cane. For instance, we have some indication from a study of more than 600 fertilizer experiments, that a higher standard error is more often associated with the lesser than with the greater amounts of fertilizer which were compared in these tests. We also recognize the fact that in variety testing the error for different canes may vary considerably; hence, one variety with a high error may give to an experiment such a large generalized error that the superiority of other varieties (with lower errors) will be unidentified. This apparent failure of the analysis of variance to identify a significant relationship, when the standard errors of the treatment yields are not homogeneous, may be illustrated with the following example of 3 Treatments (A, B, X) which were tested in 6 Blocks:

LAYOUT WITH PLOT YIELDS (T.C.A.)						SET-UP FOR TOTALS				
I		II		III		Block	A	B	X	Block totals
A	74	X	84	B	76	I.....	74	77	68	219
B	77	B	80	A	72	II.....	77	80	84	241
X	68	A	77	X	80	III.....	72	76	80	228
A	71	X	60	B	74	IV.....	71	72	74	217
B	72	B	77	A	72	V.....	72	77	60	209
X	74	A	72	X	74	VI.....	72	74	74	220
IV		V		VI		Treatment totals..	438	456	440	1334

Averages	(T.C.A.)
"A"	73 $\mp$ .9
"B"	76 $\mp$ 1.1
"X"	73 $\mp$ 3.5



## ANALYSIS OF VARIANCE

Source	Degrees of freedom	Sum squares	Mean square	"F"		Remarks
				Found	Required	
Blocks .....	5	201	....	....	....	....
Treatments .....	2	33	16.5	.73	4.10	Not significant
Error .....	10	226	22.6	....	....	....
Total.....	17	460				

This analysis indicates no effect of treatment.

Yet when these same data are examined by Student's method, we find that "B" is definitely superior to "A," viz.:

Difference						
"A"	"B"	"B" over "A"	d	d <sup>2</sup>		
74	77	+ 3	0	0	$SD = \sqrt{\frac{10}{6}} = 1.29$ $"Z" = \frac{3.0}{1.29} = 2.32$	
77	80	+ 3	0	0		
72	76	+ 4	1	1		
71	72	+ 1	2	4		
72	77	+ 5	2	4		
72	74	+ 2	1	1		
Totals .....	438	456	+18	10	n = 6	
Average .....	73	76	+ 3			

Odds = 560 to 1, and  
therefore highly sig-  
nificant.

Similarly, using Fisher's "t" value to indicate significance we would have

$$SEd = \sqrt{\frac{10}{30}} = .58 \quad t = \frac{\text{Difference}}{SEd} = \frac{3.0}{.58} = 5.2$$

and "t" required for n-1 or 5 degrees of freedom is only 2.57. Thus the difference found (3 T.C.A.) in favor of "B" is a highly significant one.

However, it is well to point out that unless there is a sound and logical reason for the high error of a Treatment average, low errors in the same test may also be due to chance and hence be unreliable. That is why the generalized error from the analysis of variance may be a safer figure to use than separately selected individual-treatment errors; and if perchance it appears that there are real differences within a test, there is no good reason why they should remain hidden, for it is still possible to make use of other statistical measures to determine the reliability of differences between the average yields.

## SUMMARY

In the preceding pages we have attempted to show by actual example how the results from field experiments can be set up and examined for evidence that the measured yield differences between treatments are really an effect of the treatments and not more likely due to chance. The use of a statistical measure as simply one more tool in the hands of the field investigator is presented in just that sense, and little effort is made to expound the basic theories of statistical analyses which

has been so aptly done by such modern leaders as "Student," Fisher, Yates, Love, Snedecor, Goulden, and others in their various publications.\*

Of especial interest because it has made possible the intelligent use of factorial experiments, the "analysis of variance" method as developed by Fisher has been quite adequately exemplified. Its utilization for identifying the contributory causes of the total "error" in an experiment should result in our placing still more emphasis on the experimental plan before the field test is actually installed.

Finally, plans and analyses for the simpler forms of factorial experiments have been offered as a suggested improvement in our experimental technique which heretofore has confined itself to the testing of single issues in separate field tests. The greater precision which is possible in securing yield effects and in estimating their significance, should make these factorial experiments doubly attractive to the plantation agriculturist.

#### *Addenda:*

For reference, and as examples of acceptable arrangements for "Grade A" experiments, we offer the following designs for the more simple Factorial Experiments:

No. 1—For a  $2 \times 2$  Factorial, e.g.  $\left. \begin{array}{l} 2 \text{ Varieties (A, X) at 2 Amounts of Nitrogen;} \\ 4 \text{ Treatments—7 Blocks—28 Plots.} \end{array} \right\}$

A1	X1	A2	X2
A2	X2	A1	X1
X2	A2	X1	A1
X1	A1	X2	A2
A1	X1	A2	
A2	X2	A1	
X2	A2	X1	
X1	A1	X2	

#### PREFERRED ANALYSIS OF VARIANCE

Source	Degrees of freedom	"F" required
Blocks .....	6	....
Varieties .....	1	4.41
Nitrogen .....	1	4.41
Interaction (V $\times$ N) ..	1	4.41
Error .....	18	....
Total .....	27	

\* References which were freely consulted during the preparation of this discussion, and with which the student of statistical methods for field experiments should be familiar, are as follows:

1. Fisher, R. A., "The Design of Experiments" published by Oliver & Boyd, London, England.
2. Goulden, C. H., "Methods of Statistical Analysis" published by John Wiley and Sons, Inc., New York.
3. Love, H. H., "Application of Statistical Methods" published by Commercial Press, Shanghai, China.
4. Snedecor, G. W., "Statistical Methods" published by the Collegiate Press, Ames, Iowa.
5. Yates, F., "The Design and Analysis of Factorial Experiments" published by The Imperial Bureau of Soil Science, Harpenden, England.

No. 2—For a  $2 \times 3$  Factorial, e.g.  $\left\{ \begin{array}{l} 2 \text{ Varieties (A, X) at 3 Amounts of Nitrogen;} \\ 6 \text{ Treatments — 5 Blocks — 30 Plots.} \end{array} \right.$

A1	X1	A3	X3	A1
X2	A2	X2	A2	X2
A3	X3	A1	X1	A3
X3	A3	X1	A1	X3
A2	X2	A2	X2	A2
X1	A1	X3	A3	X1

#### PREFERRED ANALYSIS OF VARIANCE

Source	Degrees of freedom	"F" required
Blocks .....	4	....
Varieties .....	1	4.35
Nitrogen .....	2	3.49
Interaction (V $\times$ N) ..	2	3.49
Error .....	20	....
Total .....	29	

No. 3—For a  $3 \times 2$  Factorial, e.g.  $\left\{ \begin{array}{l} 3 \text{ Varieties (A, X, B) at 2 Amounts of Nitrogen;} \\ 6 \text{ Treatments — 5 Blocks — 30 Plots.} \end{array} \right.$

A1	B2	A1	B2	A1
X2	X1	X2	X1	X2
B2	A1	B2	A1	B2
A1	B2	A1	B2	A1
X1	X2	X1	X2	X1
B2	A1	B2	A1	B2

#### PREFERRED ANALYSIS OF VARIANCE

Source	Degrees of freedom	"F" required
Blocks .....	4	....
Varieties .....	2	3.49
Nitrogen .....	1	4.35
Interaction (V $\times$ N) ..	2	3.49
Error .....	20	....
Total .....	29	

No. 4—For a  $4 \times 2$  Factorial, e.g.  $\left\{ \begin{array}{l} 4 \text{ Varieties (A, B, C, X) at 2 Amounts of Nitrogen;} \\ 8 \text{ Treatments — 4 Blocks — 32 Plots.} \end{array} \right.$

A1	X2	B2	C1
B2	C1	A1	X2
C2	B1	X1	A2
X1	A2	C2	B1
A1	X2	B2	C1
B2	C1	A1	X2
C2	B1	X1	A2
X1	A2	C2	B1

#### PREFERRED ANALYSIS OF VARIANCE

Source	Degrees of freedom	"F" required
Blocks .....	3	....
Varieties .....	3	3.07
Nitrogen .....	1	4.32
Interaction (V $\times$ N) ..	3	3.07
Error .....	21	....
Total .....	31	

No. 5—For a  $3 \times 3$  Factorial, e.g. {3 Varieties (A, X, B) at 3 Amounts of Nitrogen;  
9 Treatments—4 Graeco-Latin Squares—36 Plots.

X1	B2	A3	I
A2	X3	B1	
B3	A1	X2	
B2	A3	X1	II
A1	X2	B3	
X3	B1	A2	
X3	A1	B2	III
B1	X2	A3	
A2	B3	X1	
A1	B2	X3	IV
B3	X1	A2	
X2	A3	B1	

# PREFERRED ANALYSIS OF VARIANCE

Source	Degrees of freedom	"F" required
G. L. Squares.....	3	....
Varieties .....	2	3.40
Nitrogen .....	2	3.40
Interaction (V $\times$ N) ..	4	2.78
Error .....	24	....
Total .....	35	

No. 6—For a 3-Factor, Complex experiment: {N, P, K, each at 2 Levels (1, 2);  
8 Treatments—4 Blocks—32 Plots.

A	C	B	D	C	A
J	L	K	M	L	J
K	M	J	L	M	K
B	D	A	C	D	B
D	B				
M	K				
L	J				
C	A				

# PREFERRED ANALYSIS OF VARIANCE

Source	Degrees of freedom
Blocks .....	3
Treatments* .....	7
Error .....	21
Total .....	31

Identity	Treatment
A	= N1 P1 K1
B	= N1 P2 K1
C	= N1 P1 K2
D	= N1 P2 K2

Identity	Treatment
J	= N2 P1 K1
K	= N2 P2 K1
L	= N2 P1 K2
M	= N2 P2 K2

\* This will be partitioned to furnish 1 degree of freedom each for the following:

N    P    K  
NP   NK   PK  
NP'K

A PARTIAL TABLE OF VALUES OF "F" AND "t" CORRESPONDING TO THE 5 PER CENT LEVEL OF SIGNIFICANCE  
(Abstracted from Snedecor's\* computations from Fisher's tables)

n2	Values of "F"						Value oft	Values of "F"						Value oft
	n1	1	2	3	4	5		n1	1	2	3	4	5	
3	10.13	9.55	9.28	9.12	9.01	8.94	3.18	18	4.41	3.55	3.16	2.93	2.77	2.66
4	7.71	6.94	6.59	6.39	6.26	6.16	2.78	19	4.38	3.52	3.13	2.90	2.74	2.63
5	6.61	5.79	5.41	5.19	5.05	4.95	2.57	20	4.35	3.49	3.10	2.87	2.71	2.60
6	5.99	5.14	4.76	4.53	4.39	4.28	2.45	21	4.32	3.47	3.07	2.84	2.68	2.57
7	5.59	4.74	4.35	4.12	3.97	3.87	2.37	22	4.30	3.44	3.05	2.82	2.66	2.55
8	5.32	4.46	4.07	3.84	3.69	3.58	2.31	23	4.28	3.42	3.03	2.80	2.64	2.53
9	5.12	4.26	3.86	3.63	3.48	3.37	2.26	24	4.26	3.40	3.01	2.78	2.62	2.51
10	4.96	4.10	3.71	3.48	3.33	3.22	2.23	25	4.24	3.38	2.99	2.76	2.60	2.49
11	4.84	3.98	3.59	3.36	3.20	3.09	2.20	26	4.22	3.37	2.98	2.74	2.59	2.47
12	4.75	3.88	3.49	3.26	3.11	3.00	2.18	27	4.21	3.35	2.96	2.73	2.57	2.46
13	4.67	3.80	3.41	3.18	3.02	2.92	2.16	28	4.20	3.34	2.95	2.71	2.56	2.44
14	4.60	3.74	3.34	3.11	2.96	2.85	2.15	29	4.18	3.33	2.93	2.70	2.54	2.43
15	4.54	3.68	3.29	3.06	2.90	2.79	2.13	30	4.17	3.32	2.92	2.69	2.53	2.42
16	4.49	3.63	3.24	3.01	2.85	2.74	2.12	40	4.08	3.23	2.84	2.61	2.45	2.34
17	4.45	3.59	3.20	2.96	2.81	2.70	2.11	50	4.03	3.18	2.79	2.56	2.40	2.29

Footnote:

n1 = degrees of freedom associated with "Treatment", mean square } when determining "F", required.  
n2 = degrees of freedom associated with "Error", mean square }  
n2 = degrees of freedom for Error mean square where using "t".

\* "Statistical Methods" by G. W. Snedecor (by permission).

## ***Pythium* Root Rot of Sugar Cane in Louisiana**

(A Review by C. W. CARPENTER)

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A comprehensive account of *Pythium* root rot disease of sugar cane, as it concerns the Louisiana sugar cane industry, embodying the results of exhaustive studies and much thorough work since 1924, by Rands and Dopp,\* was published in October 1938. The virtual bankruptcy of the Louisiana sugar industry in the period 1923 to 1926 is attributed by the authors to mosaic disease, red rot and root rot. The industry recovered with the introduction of mosaic-tolerant POJ varieties; however, some of these varieties later proved susceptible to red rot and root rot. The susceptible canes were replaced by the Coimbatore canes, Co. 281 and Co. 290, and varieties bred at Canal Point, "... under the coordinated sugar cane breeding and testing program of the United States Department of Agriculture, in cooperation with the Louisiana Agricultural Experiment Station and the American Sugar Cane League." The susceptibility of the POJ varieties and some of the recently propagated hybrids to root rot indicated the need for more knowledge of root rot, and of the nature of resistance to this disease, in order to maintain or improve yields.

Previous reports by Edgerton and his co-workers, and Rands and his associates, have shown that the most important root disease of sugar cane in Louisiana is caused by *Pythium arrhenomanes*. This species is now considered identical with the *Pythium* species causing root rot in Hawaii, reported in 1920, first under the name of *P. butleri*, later revised to *P. aphanidermatum* and *P. graminicolum*, in conformance with the progress of critical studies of closely related species by various investigators. It was not considered advisable to import cultures of parasitic cane fungi to make comparative studies in Hawaii. In some of the taxonomic studies conducted by Rands and his associates and in experiments, a culture of the cane *Pythium* from Hawaii was included for comparisons of morphological characters and pathogenicity. *P. arrhenomanes* has been definitely identified thus far in Hawaii, the Philippine Islands, Mauritius, Canada and the United States, in the latter two countries as a cause of root rot disease of maize and cereals.

The fungus *P. arrhenomanes* (more recently referred to in our reports as *P. graminicolum*) is widely distributed in the cultivated soils of Hawaii, if indeed it is not ubiquitous throughout local cane lands. In the current program of testing new varieties and hybrids of local propagation, *Pythium arrhenomanes* promptly eliminates the more susceptible varieties from further consideration. It is safe to predict that no variety commercially susceptible to *Pythium* root rot will be recommended for spreading on account of its performance in our variety and regional testing stations. *Pythium* root rot apparently will always remain a hazard to the indiscriminate spreading of canes of unknown resistance.

Rands and Dopp advance the tentative hypothesis that the strains of *P. arrhenomanes* recently isolated in Louisiana may be more virulent than those isolated in

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\* Rands, R. D. (Senior Pathologist) and Dopp, Ernest (Assistant Pathologist). *Pythium* Root Rot of Sugarcane. Division of Sugar Plant Investigations, Bureau of Plant Industry, U. S. Department of Agriculture, Technical Bulletin 666, 1938.

1932 and 1933, indicating possible adaptation to the newer varieties of cane. They state that evidence of physiologic specialization of the *Pythium* sp. and the influence on yields show root rot to be a dynamic rather than a static factor in sugar cane production. According to Rands and Dopp (p. 49): "... the greatest danger from physiologic specialization of *Pythium arrhenomanes* would appear to be in connection with the less resistant varieties, which thus far unfortunately have predominated among the many new seedling selections otherwise most promising for commercial use."

The importance of *Pythium* root rot to cane production in Hawaii when varieties are naturally susceptible or become so as a result of a modified soil environment, and the potential hazard of the disease to future crops, should it be a fact that more virulent strains of the parasite are developing by a process of adaptation to our resistant crop canes, are sufficient reasons to quote in full the summary of this valuable contribution to the study of the root rot problem.

Since the failure in 1923 to 1926 of the old noble varieties of sugarcane in Louisiana, which was due to combined damage from mosaic, red rot, and the so-called root disease, the last-mentioned trouble has continued to be a serious problem; this despite the restoration of the industry from the introduction of somewhat more resistant and vigorous hybrid canes. On the latter, root rot is obviously the most important factor in the root-disease complex, even in exceptional cases when the symptoms approach those characteristic of the condition on the old varieties.

An apparent increase in the severity of root rot on certain of the newer varieties following their widespread and continued cultivation has emphasized the need for fundamental knowledge about the disease as a necessary basis for determining the resistance of new seedling selections and securing and maintaining further yield improvement.

Root rot was noted to be causing widespread damage in Louisiana as early as 1908, when red rot of the seed cane was also reported. Combined damage from the two diseases during the period 1910-20 reduced State-wide average yields by 23 percent, and the subsequent mosaic epidemic another 30 percent, which brought the industry to virtual bankruptcy. Varietal introductions by the Department and cooperating agencies (Louisiana Agricultural Experiment Station and the American Sugar Cane League) have gradually restored production to approximately that of the earlier long-period level of 1888-1907.

On certain of the presently grown and moderately susceptible varieties, such as Co. 281 and C. P. 28/19, root rot is usually manifested merely by unthrifty appearance, deficient and delayed tillering (suckering), and closing in of the rows. During occasional bad root rot years yellowing of the leaves, severe wilting, and death of young plants may result in poor stands and virtual crop failure on heavy clay soils, due to complete destruction of roots on both seed cuttings and young shoots.

As indicated in preliminary reports, *Pythium arrhenomanes* Drechsler was found to be the principal cause of the root rot. Although during the past quarter century root-disease epidemics have been reported from most sugarcane-producing countries, this particular fungus has been identified only from Hawaii, the Philippine Islands, Mauritius, Canada (where it attacks cereals), and the United States.

Twelve additional species of *Pythium* and several other fungi were isolated from decaying roots obtained in surveys of the sugar- and sirup-producing sections of the Gulf States. They were most numerous in Louisiana where the roots had either been injured by the gnawing of minute soil fauna or were weakened by red rot of the cuttings or some unfavorable soil condition. Infection tests conducted under a wide range of environmental conditions in the greenhouse were negative to the extent of development of a general root rot characteristic of *P. arrhenomanes*. They also showed no tendency to act as secondary invaders to the latter fungus. However, under the predisposing influence of dilute concentrations of a soil toxin (salicylic aldehyde) severe root rot and appreciable reduction of plant weight were caused by several of these miscellaneous species, particularly *P. dissotocum* and *P. graminicolum*. These results in

conjunction with the survey records suggest that only under very abnormal conditions may any of these species contribute to an important extent in the destruction of sugarcane roots.

Physiologic specialization and, to some extent, varietal adaptation of *Pythium arrhenomanes* in the Louisiana sugar district have been revealed by greenhouse inoculation experiments with more than 200 isolates of this species obtained in root rot surveys of representative plantations. Significant differences in average virulence of the isolates were found to occur between different plantations or localities, as well as between an earlier (1927-31) and a more recent (1935-36) survey.

Since the latter finding could not readily be explained on the basis of attenuation, resulting from prolonged maintenance in artificial culture of the early collection, actual increase in average virulence of the fungus during the period of 5 to 7 years separating the surveys is tentatively assumed to have occurred. This is conceivably due in part at least to segregation and multiplication of certain biotypes brought about by general adoption of more resistant varieties, which permitted survival of only the more virulent or adaptable components of the earlier population of the fungus.

A serious decline in yield of the susceptible P. O. J. 234 in relation to the highly resistant Co. 290 and C. P. 807 varieties in replicated agronomic yield comparisons during the past 8 years has been associated with apparent increase in root rot severity, and possibly reflects in part at least the above-found increased virulence of the *Pythium*.

No increase in root rot of resistant varieties has been observed, although one isolate of the *Pythium* was found capable of seriously damaging the Co. 290 in greenhouse tests. However, this may represent merely a chance variant rather than a specialized subpopulation of more virulent forms in the fields.

Physiologic specialization of *Pythium arrhenomanes* and its potential influence on yields show that root rot must be looked upon as a dynamic rather than a static factor, as hitherto considered in relation to sugarcane production. Therefore root rot-resistance tests of new seedling selections necessitate prior artificial infestation of the soil with a collection of the most virulent locally known cultures of the fungus.

The apparent degree of resistance or susceptibility in field tests of well-known varieties, representing the recognized species of sugarcane, is given. Most noble varieties (*Saccharum officinarum*) were found to be highly susceptible, while the Chinese canes (*S. sinense*) and the wild sugarcane (*S. spontaneum*) were highly resistant. Two Indian varieties of *S. barberi* occupied an intermediate position.

F<sub>1</sub> hybrids from crosses between the susceptible, noble, and resistant wild cane were usually resistant, but successive backcrossing to the noble parent ("nobilization") to secure commercial qualities gave increasing susceptibility in the few seedlings studied.

Since most elite breeding canes possess extremely complex inheritance, an important object of the Department's coordinated sugarcane breeding and disease-testing program is to discover more suitable parental combinations that will increase the chances of securing superior resistant varieties without increasing to prohibitive proportions the total number of seedlings to be tested.

Tentative root rot ratings on 111 first-year or later agronomic selections revealed nearly one-half to be resistant. If one or more of these should be found to combine the indispensable other qualities, especially early maturity, hazards from use of the presently available root rot-resistant varieties may be greatly minimized.

Among the present commercial varieties in Louisiana, Co. 290, C. P. 807, C. P. 28/11, and C. P. 29/116 are classed as resistant to root rot and also possess sufficient vigor for planting on the mixed and heavy soils. However, plantings of C. P. 807 have already been greatly diminished because of too great susceptibility to red rot. C. P. 29/320 has not been seriously damaged by root rot, but has been reported to be susceptible to red rot. Co. 281 and C. P. 28/19 are susceptible to root rot and ordinarily succeed only in light, well-drained soils.

Detailed studies confirmed the conclusions of other investigators that high-winter rainfall and low spring temperatures greatly accentuate the damage to fall-planted cane. Summer planting of C. P. 28/19, when it must be grown on heavy soils, was found to prevent the serious losses in yield and the practical crop failure sometimes experienced with regular October plantings.

In controlled soil temperature tanks root rot was worst at 65° to 68° F., and became progressively less serious with increase in temperature to 97° which is past optimum for cane



growth. The effect of a more virulent strain of the *Pythium* was characterized by greater damage, particularly at intermediate and high temperatures, while the use of a more resistant variety tended to suppress the disease, particularly at these temperatures.

Greater severity of root rot on mixed and heavy clay root-rot soils emphasize the need for better drainage, deep preparation by tractors, and other measures to prevent practical water-logging during periods of prolonged rainfall. The greatly accentuating effect on root rot of toxic materials possibly accumulating under such deficient aeration has been indicated by greenhouse experiments.

Increased root rot of plant cane has been noted to result apparently from excessive nitrogen fertilization of the crop furnishing the seed.

The comparative unimportance of root rot on muck and peat soils is apparently ascribable (among other things) to their greater biological activity and possible antibiotic effect on spread of the *Pythium*.

Improvement of the physical, chemical, and biological conditions of the root rot soils by continued plowing under of all cane trash and by moderate applications of factory filter-press cake or stable manure, when also accompanied by good drainage, has markedly reduced root-rot damage of susceptible varieties.

Attempts to discover a soil treatment or other direct methods for control of root rot that would not be prohibitive in cost have been unsuccessful.

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## Influence of Potash Fertilization Upon the Production and Composition of Dry Matter

By R. J. BORDEN

Investigators continue to seek ways and means to make intelligent use of plant composition figures to guide their recommendations of specific fertilizers to meet apparent plant food deficiencies in cropped soils. Only a fair amount of success has been obtained because of the many involved relationships with plant composition. One of the factors which may be concerned is apparent in the results secured from a study recently completed, which shows the effects of potash fertilization upon the production of dry matter and its potash composition and total uptake, when the crop is harvested at different stages of maturity.

### *Soils:*

Eight soils which were used in this study were obtained from Ewa Plantation Company. They all have an adequate supply of available phosphate and none are what we would consider to be greatly deficient in available potash. Briefly they are described by the following summary:

Field No.	Origin	Color	Texture	Available K20 (p.p.m.)	pH
B1	Residual	Dark red	Silty loam	250	7.2
B2	"	"	"	280	7.3
B3	"	"	"	120	7.2
B4	"	"	"	370	7.2
18A	Alluvial	Dark reddish-brown	Silty clay loam	120	7.2
18B	Marine sedimentary	Yellowish-red brown	"	110	8.2
25D	"	"	"	170	7.4
29	"	"	"	280	8.2

### *Procedure:*

After thorough preparation, standard Mitscherlich pots were filled with these soils, and three series with two potash differentials for each were provided in triplicates, i.e., 9 pots were adequately fertilized, each with 1.5 grams of K20 from sulphate of potash, and 9 pots were given no potash fertilizer. All pots were planted with Sudan grass, 40 plants being allowed to develop in each pot. Nitrogen and phosphate fertilization, and all conditions during the ensuing growth periods were similar.

Series I was harvested 50 days after planting, at a time when the crop was still growing and definitely immature. Series II was harvested at 70 days, when the plants were considered to be fully mature, while series III was allowed to become overmature and was not taken off until 90 days. Dry weights were secured from each pot at harvest, both the weight of roots and the weight of leaves and stems (with seed) being secured. Samples of all dry material were analyzed by the Chemistry department for total K20 content. Thus it is possible to calculate the

amounts of potash taken up in the dry matter produced and to determine the significant relationships.

*The Average Dry Weights:*

The effect of potash on the production of dry matter of stems and leaves and also of roots may be summarized for all 8 soils as follows:

	— Grams dry weight harvested —	
	With potash	Without potash
Series I—(50 days):		
Stems and leaves.....	905.4	890.0
Roots .....	187.2	188.1
Total dry weight.....	1,092.6	1,078.1
Series II—(70 days):		
Stems and leaves.....	1,299.1	1,195.7
Roots .....	256.4	238.8
Total dry weight.....	1,555.5	1,434.5
Series III—(90 days):		
Stems and leaves.....	1,311.8	1,284.0
Roots .....	266.4	273.1
Total dry weight.....	1,578.2	1,557.1
Dry weight total—all series.....	4,226.3	4,069.7

A small but highly significant difference (average 6.5 grams, with odds of 500 to 1) is found in the dry-weight totals which is apparently the effect of the potash fertilization. This effect is chiefly due to the increase in the aboveground part of the crop rather than to the roots. Nevertheless, a definite positive relationship ( $r = .78 \pm .04$ ) can be shown to exist between the weights of stems and leaves and the root weights from the 48 pots used in this study.

When the three series are studied separately we find that the differences between the total dry weights which favor the crops receiving potash, both in the immature series I and in the overmature series III, are not significant; the average differences of 1.8 grams and 2.6 grams respectively might easily occur by chance once in four times. On the other hand, the average difference of 15.1 grams in series II carries odds of better than 1000 to 1. This indicates that there may be some relationship between the effect of potash fertilization on the production of dry matter and the stage of the crop's development at the time it is harvested. If this indication can be reliably substantiated, it will need serious consideration.

A still further "break down" of the harvest data indicates that the differences in the average dry weights between the two treatments within each series also vary with the individual soils that were used. This is seen in the accompanying table. The reason for the negative signs in series III is not clear; a very small amount of seed shattering was not believed to be large enough to seriously affect these weights.

DIFFERENCES IN TOTAL DRY WEIGHTS (K20 OVER NO K20)  
(FROM AVERAGES OF 3 POTS OF EACH TREATMENT)

Soil	Series I Grams	Series II Grams	Series III Grams
B1	— .6	+23.5*	+ 5.1
B2	— 1.3	+ 4.0	+14.1*
B3	+ 4.3	+21.6*	+ 6.5
B4	+ 2.5	+14.8*	—12.0
18A	+11.7*	+17.3*	+ 4.3
18B	+ 6.1	+26.0*	+13.4*
25D	— 2.1	+10.0*	— 4.1
29	— 6.1	+ 3.8	— 6.2

(\* = These differences are significant, i.e., greater than  $2 \times \text{SED.}$ )

With but a few exceptions, none of which are significantly less, however, both treatments on all 8 soils show increased dry weights in both their aboveground portions and their roots, with their increased age at harvest. These increases are large and definitely significant for all 70-day harvests over their respective 50-day crop, but further gains for the 90-day harvests are smaller, and in those cases where potash was supplied are not significantly different.

AVERAGE GAIN IN TOTAL DRY WEIGHT—GRAMS

Treatment	For 70-over 50- day harvest	For 90-over 70- day harvest
With potash .....	57.8	2.6 (not significant)
Without potash .....	44.5	15.3

*Potash in Dry Matter:*

Both the percentage of potash in the dry matter and the total amount taken up by the crop were definitely influenced by the potash applications.

The percentage of K20 was substantially greater in the immature crops than in either the mature or overmature plants. In the dry matter of stems and leaves, the per cent K20 decreased directly with age of harvest. In the roots, the percentage of potash dropped significantly in all 16 comparisons of series II with series I, but it then increased significantly in all comparisons of series III with series II. This apparent return of potash to the roots after 70 days growth is most interesting. Differences in the percentage of K20 between the plants of the two treatments were consistently less as the harvests were delayed.

A summary of the per cent K20 found in the dry matter, averaged for all 8 soils appears as follows:

	—Per cent of K20—		Difference (K20 over no K20) Per cent
	With potash	Without potash	
Series I—(50 days):			
Stems and leaves.....	2.008	.994	+1.014
Roots .....	.845	.422	+ .423
Total dry weight (true average)....	1.763	.857	+ .906
Series II—(70 days):			
Stems and leaves.....	1.346	.723	+ .623
Roots .....	.375	.226	+ .149
Total dry weight (true average)....	1.184	.644	+ .540
Series III—(90 days):			
Stems and leaves.....	1.104	.545	+ .559
Roots .....	.494	.374	+ .120
Total dry weight (true average)....	.991	.514	+ .477

In these total dry weights, we find a significant decrease to an extent of 32 and 25 per cent respectively for the potash and no potash treatments in the percentage potash composition of the plants harvested at 70 days over the immature 50-day series, and a further 16 and 20 per cent decrease respectively for plants with and without potash fertilization, when the harvest is delayed from 70 to 90 days. This change in the percentage composition of potash which occurs with the stage of maturity is an important factor that will need to be considered when comparisons are being made from percentage data in plant composition studies.

The total potash recovered in the dry matter was quite definitely influenced by the potash applications. The amounts found in the total dry weight are not significantly different for the immature and mature plants but they are definitely less in the overmature crop. In the stems and leaves, there is less potash in the overmature (90-day) plants but the roots of these plants contain a larger amount than their respective 70-day series.

The total amount of potash recovered from the dry matter harvested from all 8 soils indicates some potash losses in the plant material which became greater as the harvest was delayed. This may be seen from the following summary. The apparent build-up of potash in the roots of the overmatured plants (series III) is also shown:

	Grams of —K20 in dry matter—		Grams K20 recovered	Grams K20 not recovered from amounts added
	With potash	Without potash		
Series I—(50 days):				
Stems and leaves.....	17.717	8.464		
Roots .....	1.545	.770		
Total dry weight....	19.262	9.234	10.028	1.972
Series II—(70 days):				
Stems and leaves.....	17.468	8.670		
Roots .....	.948	.563		
Total dry weight....	18.416	9.233	9.183	2.817
Series III—(90 days):				
Stems and leaves.....	14.347	6.991		
Roots .....	1.294	1.008		
Total dry weight....	15.641	7.999	7.642	4.358

Since soil analyses after harvest indicated that less than .13 gram of available K20 remained in any pot, regardless of the amount which had originally been supplied, or the length of the growing period, it appears that some of this fertilizer which was supplied is unaccounted for, in terms of available potash. For the 50-day series, this loss amounts to approximately only 17 per cent, but for the 70- and 90-day harvests, losses of 23 and 37 per cent respectively are noted. Since our pot technique precludes the loss of any leachates we can only speculate as to where this potash has gone.

#### *Correlation:*

There was no relationship between the final dry weights and the amount of potash contained therein. For the 24 pots which received no potash the correlation

coefficient ( $r$ ) was only  $.05 \pm .13$ ; and for the soils which were fertilized with potash,  $r$  was  $.02 \pm .14$ . Apparently this is an indication of the so-called "luxury consumption" of the available potash supply.

*Summary:*

1. Small gains that were secured from potash fertilization in the production of dry matter from eight soils, which are not markedly deficient in available potash, were significant only when the crop was harvested at its optimum period of development, i.e., neither immature nor overmature.

2. Although potash fertilization increased both the percentage and the total amount of this mineral found in the dry matter at harvest, there was no correlation between potash composition and final dry weights.

3. The percentage of potash in the total dry matter decreased quite substantially as the age of harvest was delayed. This was directly true for the leaves and stems, but in the roots there was apparently a return of potash when the plants passed their full maturity.

4. The total amount of potash, that was recovered in the total dry matter harvested, decreased with the age at harvest, i.e., the amount not recovered from that which had been supplied became increasingly greater as the crop reached and then passed maturity.

5. Such effects of potash fertilization as we have discussed are quite apt to introduce a complication into any attempt to interpret potash composition figures.

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# The Growth of Plants in Water and Sand Cultures

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By J. P. MARTIN AND C. W. CARPENTER

The growth of plants in water and sand cultures was started 50 years ago or more, although in recent years it has received a great deal of study and enthusiastic publicity. In the literature this technique is referred to as soilless or tray agriculture, bath tub or tank farming, chemiculture, liquid or sand culture, and recently as hydroponics (hydro = water; ponics = made with).

Many plants are now grown experimentally in water and sand cultures and a number of commercial ventures with such methods are being made on the Mainland. The method has its limitations and in order for it to be successful various procedures have to be carefully followed. Anyone trying to grow plants for the first time by the water- or sand-culture method will experience disappointments which can only be avoided by experimenting and selecting the technique best suited for his conditions. The whole system is in many ways quite flexible and the most satisfactory results are to be obtained by trial and error. If one undertakes such a project without having had previous experience, it is suggested that plants at first be grown on a small scale so that a knowledge of the plant's requirements will be secured. Experience, after all, is the best teacher.

## PLANT REQUIREMENTS

Plant growth is governed by light, temperature, moisture, and the physical and chemical qualities of the soil, all of which go to make up the environment; a change in any one of these factors may influence the rate of growth. In soilless agriculture the light and temperature factors must be favorable for plants. The chemical composition of the nutrient solution in which the plants are to be grown is comparable to the chemical qualities of the soil solution and is of paramount importance at all times to the development of the plant.

The chemical elements essential for plant growth are: hydrogen, oxygen, carbon, nitrogen, phosphorus, potassium, calcium, magnesium, sulphur, and iron; in addition to these are the trace elements: manganese, boron, zinc, copper and perhaps others. Hydrogen is obtained chiefly from water, oxygen from water and air, while carbon is taken in by the leaves from the air as carbon dioxide. The other elements are taken from the soil. For normal growth in water cultures or in the soil these elements must be present in sufficient quantities and in available forms. A deficiency or an excess of any one element produces an abnormal growth and frequently these abnormalities prove extremely useful in determining the plant's requirements or in diagnosing physiological diseases such as nitrogen or iron deficiency, as described below.

## THE SOLUBLE PLANT FERTILIZER

In recent years, sugar cane has been successfully grown in water and sand cultures with various nutrient solutions at the Experiment Station, Hawaiian Sugar, Planters' Association. Other plants such as tomatoes, potatoes, and asparagus



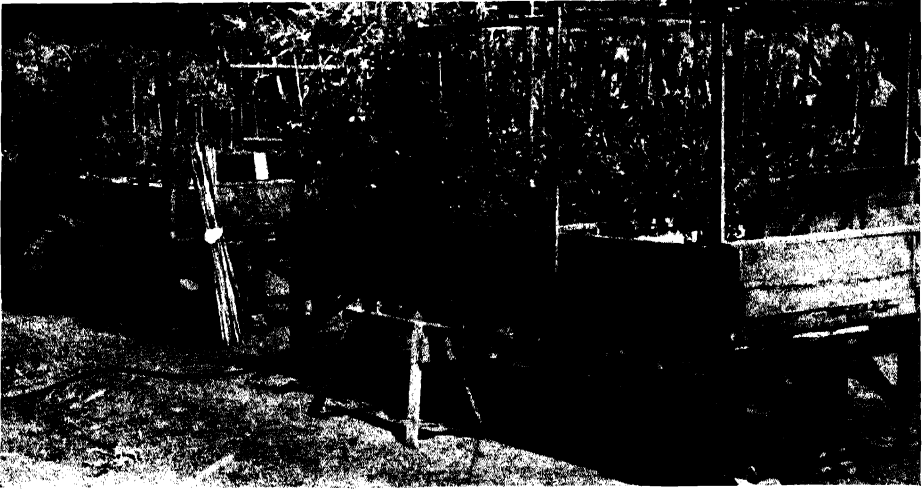


Fig. 1. Young tomato plants growing out of doors in black sand and irrigated with the Soluble Plant Fertilizer solution.



Fig. 2. Young tomato plants growing within a screened house in black sand and irrigated with a nutrient solution prepared from the S. P. Fertilizer.

which were used in special studies developed normally in sand cultures when irrigated with the nutrient solutions used for sugar cane.

With the thought in mind that a complete, soluble plant fertilizer might be useful for growing various plants without soil, as well as for stimulating the growth of potted plants, a water-soluble fertilizer was prepared, after a number of trials, from commercial chemicals. The various elements in this mixture are present in amounts equal to those in the nutrient solution which proved to be best adapted for sugar

carries both known and undetermined trace elements, has proved superior for plant growth in a number of cases to nutrient solutions prepared with chemically pure salts.

The S.P. Fertilizer is packed in air-tight containers, labeled, respectively, A, which contains the nitrogen, and B, which contains the other elements essential for growth; it may be obtained with directions for its use from the Pacific Guano and Fertilizer Company. The mixture has been prepared so that one teaspoonful each of A and B to two gallons of water gives a concentration satisfactory for plant growth. For some plants it might be advisable to make the nutrient solution slightly more acid; this is accomplished by adding dilute sulphuric acid to the tap water prior to the addition of the fertilizer; for example: if the original pH value of the tap water is 8.0, then 5 c.c. of normal sulphuric acid is usually required to bring the final pH value of the nutrient solution to 5.3 (further directions are given below).

Plants grown in sand cultures are irrigated lightly two or three times a week with the S.P. Fertilizer solution. As the plants begin to grow they are irrigated twice a week and supplied with tap water if they require additional moisture (Figs. 1 and 2).

If plants are grown in water cultures the containers are nearly filled with the solution which is renewed every ten days or two weeks. The frequency at which the solutions are changed will depend largely on the size of the plants, their rate of growth and the volume of solution in the containers. If deep containers are used some means for aerating the solutions should be provided. Details relating to aerating solutions, the size of containers, and mechanical supports for the plants are discussed below.

Tomatoes have been grown mainly in sand cultures and the yields have been quite satisfactory. Various stages of growth of the plants and the type of containers employed are shown in Figs. 1 and 2.

For potted plants the solution containing the S.P. Fertilizer is applied directly to the soil; an application once every two weeks has been found to be satisfactory for a number of the ordinary house plants but more or less frequent applications may be made according to the growth response desired.

The commercial fertilizers for soilless agriculture now on the market contain most of the elements listed above under "Plant Requirements," but the concentration of each element in one mixture may vary considerably from that in another mixture without greatly affecting the growth of the plant—in other words there is, as a rule, a margin of safety in the concentrations of each element for plant growth. A modification of the concentrations of one or more elements may be necessary to meet the requirements of some plants.

#### WATER VS. SAND CULTURES

Plants absorb the mineral nutrients from the soil solution through their roots. One important function of the soil is to accommodate a large root surface for absorption; another is to serve as a physical support or anchorage for the plant. The soil is to a greater or lesser degree self-aerating and self-draining. Plants grow well in water culture and in sand culture but some means of physical support must be provided in water culture. In the sand-culture method the quartz, beach, or black (volcanic cinders) sand provides physical support; the culture solution, added fre-

cane growth. This mixture, known as the *Soluble Plant (S.P.) Fertilizer*, which quently or continuously by mechanical means, provides the nutrients. If coral sand is to be used as a medium it should first be thoroughly washed with tap water and then rinsed with acidified water before attempting to grow plants in it. Coral sand selected from a locality well removed from the ocean where it has been exposed to rains is preferable to sand taken from the ocean's edge.

The most common design for water cultures includes a water-tight tank of metal or wood with a movable screened frame which fits the top. On the screened frame is placed a layer of excelsior, shavings, or other similar inert material (the substratum) in which the seed is planted or the seedlings transplanted; the roots penetrate this layer and are constantly submerged in the culture solution. For small plants various containers such as crocks or fruit jars may be used.

The tanks may be of various materials and of various shapes and sizes according to the size and number of plants to be grown. The tank should be water tight, the material non-corrosive, or covered with a neutral and safe coating such as a pure non-oily asphalt paint. Paint with a lead or other metallic base should be avoided. Galvanized iron and redwood, unless coated as described above, are unsatisfactory. The depth of the solution required varies somewhat with the root habits of the plants to be grown. Tubers and bulbs develop in the excelsior or other substratum and only the roots penetrate into the solution; for such crops a proportionately deeper layer of substratum is needed. Relatively shallow tanks provide sufficient depth without requiring excessive quantities of nutrient solution in proportion to the surface available for plant growth.

The tray may be made to cover the tank, or it may be made slightly smaller, to slip inside and rest on supports just above the solution. Sufficient space should be provided at the end of the tray to permit inspection and changing the solution. Aeration of the solution is necessary especially if deep containers are used. In small tanks the tray may be lifted several times a day so that the roots may carry air into the solution, or in larger tank cultures mechanical stirring, circulation of the solution by pumps, or bubbling air through the solution may be used to provide adequate circulation and aeration. Mosquito larvae (wigglers) frequently develop in water cultures unless the plants are grown in a screened house or the access of mosquitoes to the culture solution is otherwise prevented. The larvae cling to the roots and are not easily eliminated when the solutions are renewed.

Sand cultures are in many ways preferable to water cultures; the plants are supported by the sand, and the aeration as well as drainage are taken care of during the irrigation. The solution may be allowed to drip slowly onto the sand, collected below the cultures and used repeatedly—the drip method. With the aid of circulating pumps the solution may be applied continuously by drip or faster irrigation. Lack of aeration frequently accompanied by stagnation of the solution and decomposition of the roots is avoided to a large extent by using sand cultures.

*Pests and Diseases:* Plants grown in soilless culture are not immune to the diseases and insect pests of the field. Since plants are concentrated in a limited area, the pests may spread easily but they are more readily observed and measures for control may be more easily applied. Plants grown under cover, e. g., in a glass house, are particularly subject to infestation with plant lice and mites. Frequent sprayings of the foliage with water aid to simulate rain and dew conditions of the

field and tend to restrict the insect infestations. Spraying with nicotine-sulfate solution and dusting with sulphur may be required occasionally to control aphids and mites. The plants may be sprayed with Bordeaux mixture if fungous diseases are troublesome.

When sand is used for three months or more the root knot nematode may attack the roots of the plants and prevent normal growth or even kill the plants. Sand infested with nematodes, after continuous growth of a susceptible plant for several months, must be sterilized by heat or chemical disinfectants before satisfactory growth of any susceptible plant can be expected. Many vegetables are particularly susceptible to nematode attack, for example: tomatoes, potatoes, etc.

*Expense:* The cost of small outfits for either water or sand cultures in the home is nominal. Commercial units are expensive and still in the experimental stage. Before the venture is attempted on a commercial scale, a site should be selected near a good market for out-of-season products which command and receive premium prices.

#### PRACTICAL USES OF SOILLESS AGRICULTURE

At the present time it may be said that soilless agriculture is an interesting hobby which produces edible or other useful products. Such culture is distinctly advantageous where neither good soil nor water are available in quantity (certain Pacific Islands). In favorable localities near large cities some commercial "tank farms" are being operated successfully by experienced growers of fancy agricultural products.

Any one who becomes interested in soilless agriculture should not be influenced by the too-enthusiastic propaganda written by laymen who eulogize tank farms and optimistically predict the ultimate substitution of tank culture for field agriculture. A statement has been made by a Mainland authority that it would require 20,000 times the annual production of phosphate in the United States to grow all of our vegetables by the tank-culture method.

For an entertaining and popular discussion of the water-culture method of growing plants the reader is referred to an article by Frank J. Taylor entitled "You Can Try It Yourself" in the *Saturday Evening Post*, August 20, 1938.

#### PREPARATION OF CULTURE SOLUTIONS (HOAGLAND AND ARNON)

A brief account of the water-culture method of growing plants, by D. R. Hoagland and D. I. Arnon of the College of Agriculture of the University of California, appeared in the C.R.E.A. (Committee on the Relation of Electricity to Agriculture) News Letter for June 1938. The authors state that many of the popular articles on the water-culture method of crop production are "grossly inaccurate in fact, and misleading in implications."

We are indebted to Hoagland and Arnon for the following formulae and directions for their use; the first formula we quote for the benefit of those who may wish to prepare culture solutions from fertilizers and chemicals of ordinary grade; the second, a more technical formula, we quote for use in schools where laboratory facilities are available.

*Preparation of Nutrient Solutions: Method A, for Amateurs*

Either one of the solutions given in Table V may be tried. The "T.C." solution may often be preferred because the ammonium salt delays the development of undesirable alkalinity. The salts are added to the water, preferably in the order given.

To either of the solutions add the elements iron, boron, manganese, zinc, and copper, which are required by plants in minute quantities. There is danger of toxic effects if much greater quantities of these elements are added than indicated later in the text.

With the exception of iron, the elements of this group are added only when the solution is first prepared or when the whole solution is changed.

TABLE V. COMPOSITION OF NUTRIENT SOLUTIONS  
(The amounts given are for 25 gallons of solution)

Salt	Grade of salt	Approx. amt. in ounces	Approx. amount in tablespoons
<i>"P.N." Solution</i>			
Potassium phosphate (monobasic)	Technical	$\frac{1}{2}$	1 level
Potassium nitrate	Fertilizer	2	4 level (of powd. salt)
Calcium nitrate	Fertilizer	3	7 level
Magnesium sulfate (Epsom salt)	Technical	$1\frac{1}{2}$	4 level
<i>"T.C." Solution</i>			
Ammonium phosphate (monobasic)	Technical	$\frac{1}{2}$	1 heaping
Potassium nitrate	Fertilizer	$2\frac{1}{2}$	5 level (of powd. salt)
Calcium nitrate	Fertilizer	$2\frac{1}{2}$	6 level
Magnesium sulfate (Epsom salt)	Technical	$1\frac{1}{2}$	4 level

It may be necessary to add the iron solution at frequent intervals; for example, once or twice a week. If the leaves of the plant tend to become yellow the reason may be lack of iron, although a yellowing or mottling of leaves can also be due to other causes.

1. Iron Solution.

Dissolve a level teaspoon of iron tartrate (iron citrate or iron sulfate can be substituted, but the tartrate or citrate are often more effective than the sulfate) in a quart of water. Add half a cupful of this solution to 25 gallons of nutrient solution each time iron is needed (once weekly, or more frequently if the plants are pale).

2. Boron Solution.

Dissolve a level teaspoon of powdered boric acid in a gallon of water. Use a pint and a half of this solution for each 25 gallons of nutrient solution.

3. Manganese Solution.

Dissolve a teaspoon of crystalline, chemically pure manganese chloride ( $\text{MnCl}_2 \cdot 4 \text{H}_2\text{O}$ ) in a gallon of water. Manganese sulfate can also be used. Dilute one part of this solution with two parts of water, by volume. Use a pint of the *diluted* solution for each 25 gallons of water.

4. Zinc Solution.

Dissolve a level teaspoon of crystalline, chemically pure zinc sulfate ( $\text{ZnSO}_4 \cdot 7 \text{H}_2\text{O}$ ) in a gallon of water. Use four teaspoons of this solution for each 25 gallons of nutrient solution.

5. Copper Solution.

Dissolve a teaspoon of chemically pure copper sulfate ( $\text{CuSO}_4 \cdot 5 \text{H}_2\text{O}$ ) in a gallon of water. Dilute one part of this solution with four parts of water; use one *teaspoon* of the *diluted* solution for each 25 gallons of nutrient solution.

*Testing and Adjusting the Acidity of Water and Nutrient Solution*

The chemicals required are:

1. Brom thymol blue indicator. This can be obtained, with directions for use, from chemical supply houses, in the form of solutions or impregnated strips of paper.

2. Sulfuric acid. Purchase a supply of three per cent (by volume) acid of chemically pure grade. (Concentrated, chemically pure sulfuric acid may be purchased and diluted to three per cent strength, *but the concentrated acid is dangerous to handle by inexperienced persons*). This three per cent acid may be further diluted with water if a preliminary test indicates that only small additions of acid are required to bring about a desirable reaction.

Adjust the acidity of the water before adding nutrient salt according to directions given.

Test the degree of acidity of a measured sample of the water (a quart, for example) by noting the color of the added indicator or test paper immersed in the solution.

A yellow color indicates the desired slight acidity (with no further adjustment necessary), green a neutral reaction, blue an alkaline reaction.

Add the dilute sulfuric acid (three per cent or less) slowly with stirring until the original green or blue color *just* changes to yellow. Do not add more acid beyond this point, since the yellow color will also persist when excessive amounts of acid are added. Record the amount of acid required.

Finally add a proportionate amount of the acid to the solution in the culture tank or vessel, having first determined how much it holds.

The reaction of the culture solution should be likewise tested from time to time and, if found alkaline, corrected by the addition with stirring of dilute sulfuric acid. If strips of indicator paper are used, the test may be performed directly in the tank, or on a small sample of the culture solution.

*Preparation of Nutrient Solutions: Method B, for Special Experimentation by Schools, etc.*

The use of distilled water and chemically pure salts is recommended. Molal stock solutions (except when otherwise indicated) are prepared for each salt, and the amounts indicated below are used.

"P.N." solution	Cc. per liter of nutrient solution
M/1 $\text{KH}_2\text{PO}_4$ potassium acid phosphate.....	1
M/1 $\text{KNO}_3$ potassium nitrate.....	5
M/1 $\text{Ca}(\text{NO}_3)_2$ calcium nitrate.....	5
M/1 $\text{MgSO}_4$ magnesium sulfate.....	2
"T.C." solution	
M/1 $\text{NH}_4\text{H}_2\text{PO}_4$ ammonium acid phosphate.....	1
M/1 $\text{KNO}_3$ potassium nitrate.....	6
M/1 $\text{Ca}(\text{NO}_3)_2$ calcium nitrate.....	4
M/1 $\text{MgSO}_4$ magnesium sulfate.....	2

To either of these solutions add the following:

(a) Iron in the form of 0.5 per cent iron tartrate solution or other suitable iron salt, at the rate of 1 cc. per liter, about once weekly or as indicated by appearance of plants (more if pale).

(b) Prepare a supplementary solution which will supply boron, manganese, zinc, and copper, as follows:

Compound	Grams dissolved in 1 liter of $\text{H}_2\text{O}$
$\text{H}_3\text{BO}_3$ .....	2.86
$\text{MnCl}_2 \cdot 4 \text{H}_2\text{O}$ .....	1.81
$\text{ZnSO}_4 \cdot 7 \text{H}_2\text{O}$ .....	0.22
$\text{CuSO}_4 \cdot 5 \text{H}_2\text{O}$ .....	0.08

Use 1 cc. of this solution for each liter of nutrient solution. This will give the following concentrations:

Element	Parts per million of nutrient solution
Boron .....	0.5
Manganese .....	0.5
Zinc .....	0.05
Copper .....	0.02

*Adjustment of Reaction During Growth of Plants*

If the culture solution should become alkaline (pH greater than 7) as a result of growing plants, make the solution slightly acid (about pH 6) by adding N/10  $\text{H}_2\text{SO}_4$  (or some other suitable dilution).

*Changes of Nutrient Solution*

As the plants begin to grow, nutrient salts will be absorbed and the acidity of the solution will change. More salts and acid may be added, but to know how much, chemical tests on the solution are required. When these cannot be made, an arbitrary procedure may be adopted of draining out the old solution every week or two, immediately refilling the tank with water, and adding salts and acid, as at the beginning of the culture. The number of changes of solution required will depend on size of plants, how fast they are growing, and on volume of solution. Distribute the salts and acid to different parts of the tank. In order to effect proper mixing, it may be well to fill the tank at first only partly full (but keep most of the roots immersed) and then after adding the salts and acid, to complete the filling to the proper level with a rapid stream of water.

In December 1938, Circular 347, entitled "The Water Culture-Method for Growing Plants Without Soil" by D. R. Hoagland and D. I. Arnon was published by the University of California. The authors have presented the principles and application of the water-culture method together with its possibilities and limitations, and have given detailed directions for growing plants by this method. This article should be extremely valuable to those interested in soilless agriculture.

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## Variation in Available Nutrients in an Uncropped Surface Soil

By R. J. BORDEN

It seems desirable to know the possible extent of the natural variation in the available soil nutrient content of a soil, so that a more reliable interpretation can be given to results of soil analyses when they are being used to guide fertilizer practices.

In a previous study (1), we have reported upon the seasonal variations in the availability of the principal nutrients in a soil while a cane crop was being grown thereon. In this present study (2) we have attempted to secure a similar picture from a soil which was maintained in bare fallow.

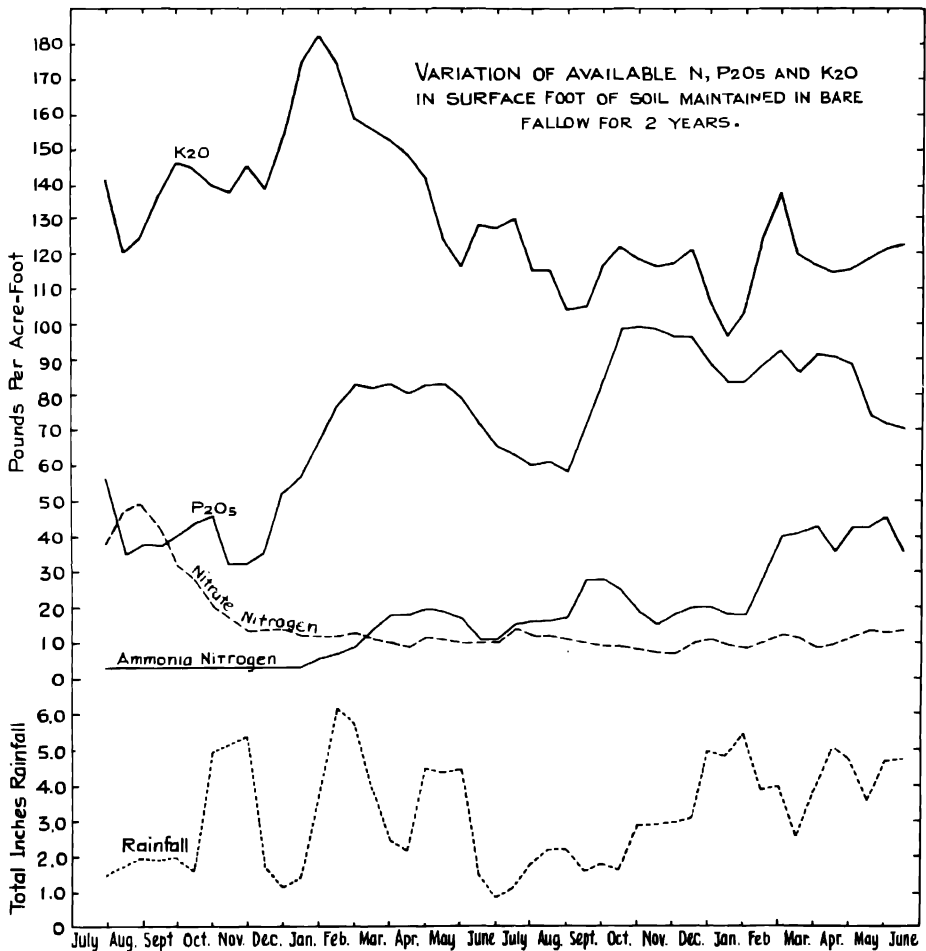


Fig. 1.

- (1) Reported in The Hawaiian Planters' Record, 41: 47-55, 1937.
- (2) Experiment Station, H.S.P.A., Project A-105—No. 82.



We selected a small area approximately 25 feet square at our Kailua substation, removed all plant growth, and maintained a clean surface soil thereon for a period of 2 years. During this period, twice each month, two separate soil sample composites from 10 auger borings each were taken from each of three soil depths: (a) 0-6", (b) 6"-12"; and (c) 12"-24". Auger holes from which soil was taken were refilled and marked with wire pins, so that each subsequent boring could be made not more than 6 or 8 inches from the previous bore. With such a procedure, the successively taken soil samples are believed to be quite comparable.

Soil samples thus taken (3) were analyzed (4) by rapid chemical methods in our soils laboratory. Results in detail are given in Table I, as pounds of available nutrients per acre in the three soil depths indicated.

A somewhat clearer picture of the variations that were found may be obtained from Fig. 1, which shows a smoothed curve (from moving averages) for the amounts of various nutrients found in the upper foot of soil, i.e., the total found in the 0-6" and the 6"-12" depths. As a matter of interest, a smoothed curve for rainfall is also included.

It is quite apparent that a variation in the available nutrient supply of this soil existed during the two-year period while it was being studied. The tabulated data indicate the range of this variability to be as follows:

For potash: from 75 lbs. on January 18, 1938 to 200 lbs. on January 18, 1937.

For phosphate: from 33 lbs. on August 18, 1936 to 107 lbs. on April 2, 1938.

For ammonia nitrogen: from less than 5 lbs. prior to February 1937 to 53 lbs. on May 5, 1938.

For nitrate nitrogen: from 6 lbs. on April 2, 1938 to 56 lbs. on August 3, 1936.

The differences in soil pH were perhaps not significant, but rainfall per half-month period varied from a half-inch to twelve inches.

Although from a strictly quantitative standpoint these ranges may appear large, the facts are not quite so disconcerting if we recognize the limitations in the practical application of soil analytical data and look at them from a qualitative or perhaps a semi-quantitative point of view. For instance: our qualitative grouping\* of soils with respect to the availability of nutrients as found by R.C.M. analyses would indicate that the potash supply during the first year's samplings varied principally *within* the "doubtful" group, reaching 200 pounds only once and never going below 100 pounds. During the second year's sampling, the potash content of the periodic sample was quite generally *within* the "low" group, and it never got above 150 pounds. Thus from a practical standpoint the analyses during both years consistently call our attention to the fact that the available potash supply was inade-

(3) Taken by various assistant-agriculturists-in-training under supervision of K. H. Berg or L. R. Smith, and Y. Yamasaki.

(4) All R.C.M. analyses by H. M. Lee.

\* Pounds per Acre-Foot of Available Nutrients by R.C.M.

Soil group	P <sub>2</sub> O <sub>5</sub>	K <sub>2</sub> O	N
Low .....	0-20	0-125	0-25
Doubtful .....	21-35	126-200	26-50
Medium .....	36-100	201-300	51-100
High .....	100+	300+	100+

quate, from which we know that potash fertilization will be necessary for successful cane growth on this soil.

In the same way, the analyses of the periodic samples show that the phosphate supply varied considerably, but that quite generally all of the differences were well *within* the "medium" phosphate grouping. Only two samples actually got into the "high" group, and these showed amounts that are only just over the border of the "medium" group upper limit, i.e., 104 and 107 pounds respectively; similarly only three samples were below the lower limit of the "medium" group, i.e., at 33, 34, and 35 pounds respectively. Hence the soil in this area is shown to have a fair amount of available phosphate but not sufficient to warrant omitting phosphate entirely from its proposed fertilizer program. Yet, it is extremely doubtful that the figures obtained could be reliably considered as actual quantitative data and evaluated as such when planning the amount of  $P_2O_5$  one would need to supply in the fertilizer to make up for the deficiency which apparently exists in the soil.

The nitrogen picture illustrates still further variations which are probably the effect of changes in the activity of soil organisms and the fate of their products. The nitrate-nitrogen status after the first few months decreased to a figure close to 10 pounds per acre, from which it seldom varied thereafter. The ammonia nitrogen, which was negligible for some six months at the beginning of the study, gradually accumulated thereafter. At no time, however, was the available nitrogen content sufficiently high to indicate that it would be feasible to make any allowance in the total supply that might be required and which might be proposed for a sugar cane crop.

Such evidence of variations, as we have recorded herewith, leads us to recognize better the limitations concerned with the use of soil analyses data, and indicates how hazardous it would be to evaluate the results on a strictly quantitative basis when deciding on the actual amounts of plant food to be supplied in the fertilizer for sugar cane.

TABLE I  
DETAIL OF SEMI-MONTHLY SAMPLES  
(All figures for nutrients are averages of two samples given as pounds per acre to depths sampled)

Date sampled	lb ammonia nitrogen—			lb nitrate nitrogen—			lb P <sub>2</sub> O <sub>5</sub> —			lb K <sub>2</sub> O—			pH—			Inches rainfall ( $\frac{1}{4}$ mo.)
	0-6"	6"-12"	12"-24"	0-6"	6"-12"	12"-24"	0-6"	6"-12"	12"-24"	0-6"	6"-12"	12"-24"	0-6"	6"-12"	12"-24"	
July 3, 1936	-5	-5	-5	16	9	7	62	37	163	106	69	100	6.5	6.6	6.7	.99
July 16	-5	-5	-5	20	11	5	21	14	18	69	37	-75	6.5	6.6	6.7	1.76
August 3	-5	-5	-5	36	20	17	17	17	20	81	56	-75	6.4	6.6	6.7	1.70
August 18	-5	-5	-5	34	18	17	18	15	25	75	37	-75	6.3	6.5	6.5	1.73
September 3	-5	-5	-5	21	18	25	21	16	18	75	44	-75	6.4	6.5	6.6	2.33
September 18	-5	-5	-5	19	16	22	23	17	15	131	44	-75	6.6	6.7	6.9	1.64
October 3	-5	-5	-5	11	9	20	27	17	22	106	37	-75	6.5	6.5	6.7	2.02
October 18	-5	-5	-5	13	15	26	35	14	18	75	37	-75	6.6	6.6	6.7	.94
November 3	-5	-5	-5	4	7	14	31	15	23	125	33	-75	6.6	6.6	6.6	12.00
November 18	-5	-5	-5	4	7	23	27	23	28	94	44	-75	6.4	6.5	6.5	2.62
December 3	-5	-5	-5	7	10	23	37	23	28	94	44	-75	6.5	6.6	6.6	1.66
December 18	-5	-5	-5	5	8	15	39	16	28	94	44	-75	6.4	6.6	6.5	.99
January 3, 1937	-5	-5	-5	5	7	13	27	23	25	131	50	-75	6.5	6.5	6.6	.83
January 18	-5	-5	-5	4	7	15	43	23	30	150	50	-75	6.6	6.5	6.6	2.62
February 3	5	4	11	4	8	13	50	35	30	125	37	-75	6.8	6.6	6.7	8.26
February 18	5	3	5	5	9	20	50	29	62	125	33	88	6.7	6.6	6.7	7.89
March 3	6	6	7	7	18	5	50	35	25	119	33	-75	6.7	6.7	6.8	2.71
March 18	11	10	18	2	4	10	50	31	30	119	33	-75	6.8	6.8	6.9	1.38
April 3	11	9	13	3	6	10	62	19	25	112	37	-75	6.8	6.8	6.9	3.45
April 17	7	4	8	5	5	12	47	31	32	106	33	-75	6.8	6.8	6.8	.50
May 3	16	11	18	6	9	4	50	39	62	87	44	-75	6.7	6.7	6.7	9.75
May 18	10	8	12	4	4	5	50	31	82	63	37	-75	6.7	6.7	6.7	2.99
June 3	3	3	3	3	4	7	47	21	25	81	33	-75	6.6	6.6	6.6	.77
June 18	7	4	9	9	5	9	39	50	30	131	37	-75	6.6	6.5	6.5	1.19
July 3	11	7	15	6	6	7	35	29	62	62	36	61	6.6	6.6	6.6	.70
July 18	9	8	14	8	6	8	35	28	58	87	36	54	6.6	6.6	6.6	1.61
August 3	6	6	12	5	5	9	29	25	56	87	36	55	6.6	6.6	6.6	3.27
August 18	12	9	15	6	6	6	47	20	34	62	36	61	6.6	6.6	6.6	1.77
September 3	10	8	14	5	5	8	35	19	28	56	37	41	6.6	6.6	6.6	1.77
September 17	27	16	30	4	3	4	50	43	98	81	44	44	6.9	7.0	6.9	1.34
October 4	12	9	13	5	5	7	57	47	93	94	44	39	6.7	6.8	6.8	2.47
October 18	6	3	8	6	5	8	49	50	93	62	39	55	6.7	6.8	6.7	1.16
November 3	14	9	22	4	4	0	47	43	113	75	39	75	6.9	7.0	6.8	5.16
November 18	7	6	14	3	3	4	50	50	105	87	45	71	6.8	6.9	6.7	2.45
December 3	12	6	12	4	6	12	47	47	85	62	40	67	6.8	6.9	6.8	1.34
December 18	15	12	14	3	10	16	48	47	80	82	44	75	6.5	6.5	6.6	5.54
January 3, 1938	9	4	9	4	5	8	40	37	69	50	37	72	6.5	6.6	6.7	8.20
January 18	6	6	7	3	4	7	35	43	62	37	38	68	6.6	6.8	6.9	.84
February 3	18	11	19	10	8	8	57	39	62	100	50	88	6.6	6.6	6.6	7.46
February 18	21	24	24	3	12	8	53	39	54	94	50	75	6.6	6.6	6.6	3.39
March 3	22	22	45	6	4	8	50	39	93	75	42	88	6.5	6.6	6.6	1.31
March 18	17	15	20	3	6	11	47	29	69	56	40	68	6.6	6.6	6.6	3.04
April 2	34	16	29	3	3	12	57	50	93	94	45	80	6.6	6.6	6.6	7.07
April 18	14	9	35	7	6	7	47	41	93	69	42	65	6.5	6.6	6.6	5.24
May 5	27	26	30	6	7	5	39	31	93	62	36	54	6.5	6.6	6.6	1.94
May 18	24	27	34	7	5	9	39	23	78	100	44	78	6.6	6.6	6.6	3.34
June 3	16	14	27	6	7	8	47	35	62	69	52	78	6.4	6.5	6.5	8.71
June 18	12	9	13	8	7	12	43	23	58	62	39	54	6.5	6.6	6.5	2.08

Note: lb N, P<sub>2</sub>O<sub>5</sub>, K<sub>2</sub>O — for 0-6", and 6"-12" depths =  $\frac{\text{p.p.m.} \times 2.5}{2}$   
for 12"-24" = p.p.m. x 2.5.

## Colorimetric Method for the Determination of Sulfate in Cane Juice

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By PAUL E. CHU and FRANCIS E. HANCE

Colorimetric methods for the determination of sulfate have been described by a few workers. These have been principally in the field of biological chemistry.

Kahn and Leiboff (6), Wakefield (10) and others isolated sulfate as benzidine sulfate. The benzidine is diazotized and the color developed with phenol in an alkaline solution.

Hubbard (5) treated solutions of benzidine sulfate with hydrogen peroxide and ferric chloride to produce yellow solutions. These were compared with standards similarly treated.

Letonoff (8) and his colleagues used Folin's amino acid reagent in conjunction with benzidine for sulfate in serums and urine.

These methods are somewhat lengthy and proved unsatisfactory as rapid chemical methods (3, 4). It was our purpose, also, to have permanent standards for comparison with the colors developed.

In the search for a method suitable for the colorimetric analysis of sulfate in cane juice, it was noted that a number of workers, Abrahamczik and Blumel (1), Giblin (2) and Kochor (7) had used sodium rhodizonate as an external indicator, and Mutschin and Pollak (9) had used the same salt as an internal indicator in volumetric methods for sulfate.

The proposed colorimetric method utilizes the color formed by sodium rhodizonate and the excess barium chloride which is used to precipitate the sulfate in the sample.

In brief, the method follows: 0.50 ml. of 0.01 N barium chloride solution is added to a measured portion of the juice sample which is placed in a tall vial (phosphate type). The contents of the vial are shaken for ten seconds, allowed to stand one-half minute, made up to 7.0 ml. with distilled water and thoroughly mixed. Three-fourths of a ml. of a freshly prepared 0.1 per cent aqueous solution of sodium rhodizonate is added and the contents again mixed to develop the color. The sodium rhodizonate forms a red solution with the excess barium. If there is no excess barium chloride, the solution is yellow.

A set of eight permanent inorganic standards, Plate I, has been prepared to cover the range of colors developed. The test solution is compared with the sulfate standards in front of a phosphate illuminator (3). A table gives the sulfate content in terms of parts per million sulfate for various aliquots of the sample which match each of the standard tubes.

It is necessary to follow precisely the proportions given in the method when using the standards described below. After exhaustive study, the concentrations and proportions of reacting substances given in the following detailed procedure were found to be most satisfactory.

EXPLANATION OF PLATE I  
COLORIMETRIC METHOD FOR THE DETERMINATION OF SULFATE IN CANE JUICE

The sealed tubes of color standards are arranged in the order of increasing sulfate content, the lowest being at the extreme left. They are placed in a wooden rack and are numbered progressively from one to eight, the lower figure denoting lower sulfate content.

Unknown solutions in open vials are placed in the intervening spaces and the whole assembly is placed in front of a standard source of illumination for comparison. Reference is made to a suitably prepared table for analytical values. The standards are made from an inorganic salt and are permanent. Full details of preparation, standardization and evaluation appear in the text.

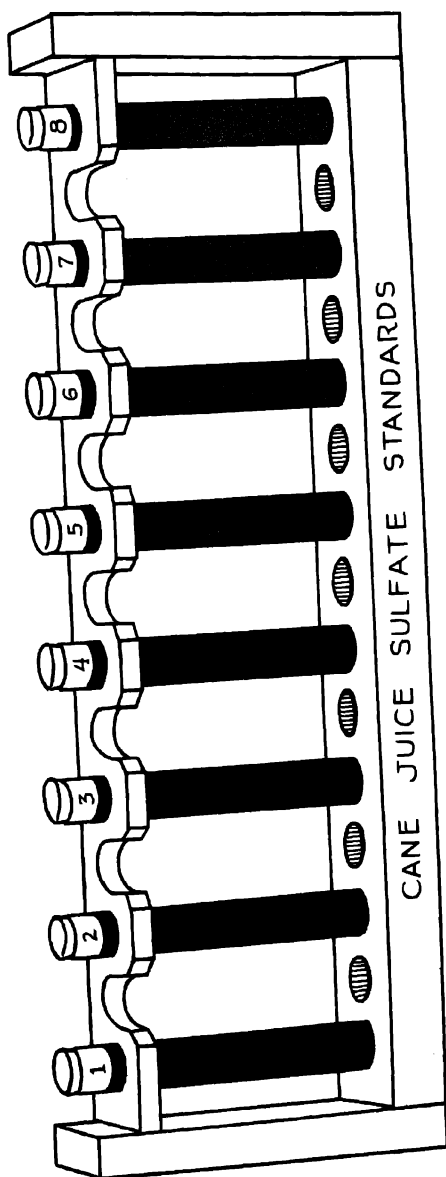


PLATE I.



*Permanent Inorganic Color Standards:*

The search for permanent soluble inorganic salts, or combinations of these, to match the colors developed in the test vials in the determination of sulfate encountered unusual difficulties because the red-colored barium rhodizonate was mixed with small crystals of white barium sulfate. This mixture produced a tinted turbidity instead of clear-colored solutions.

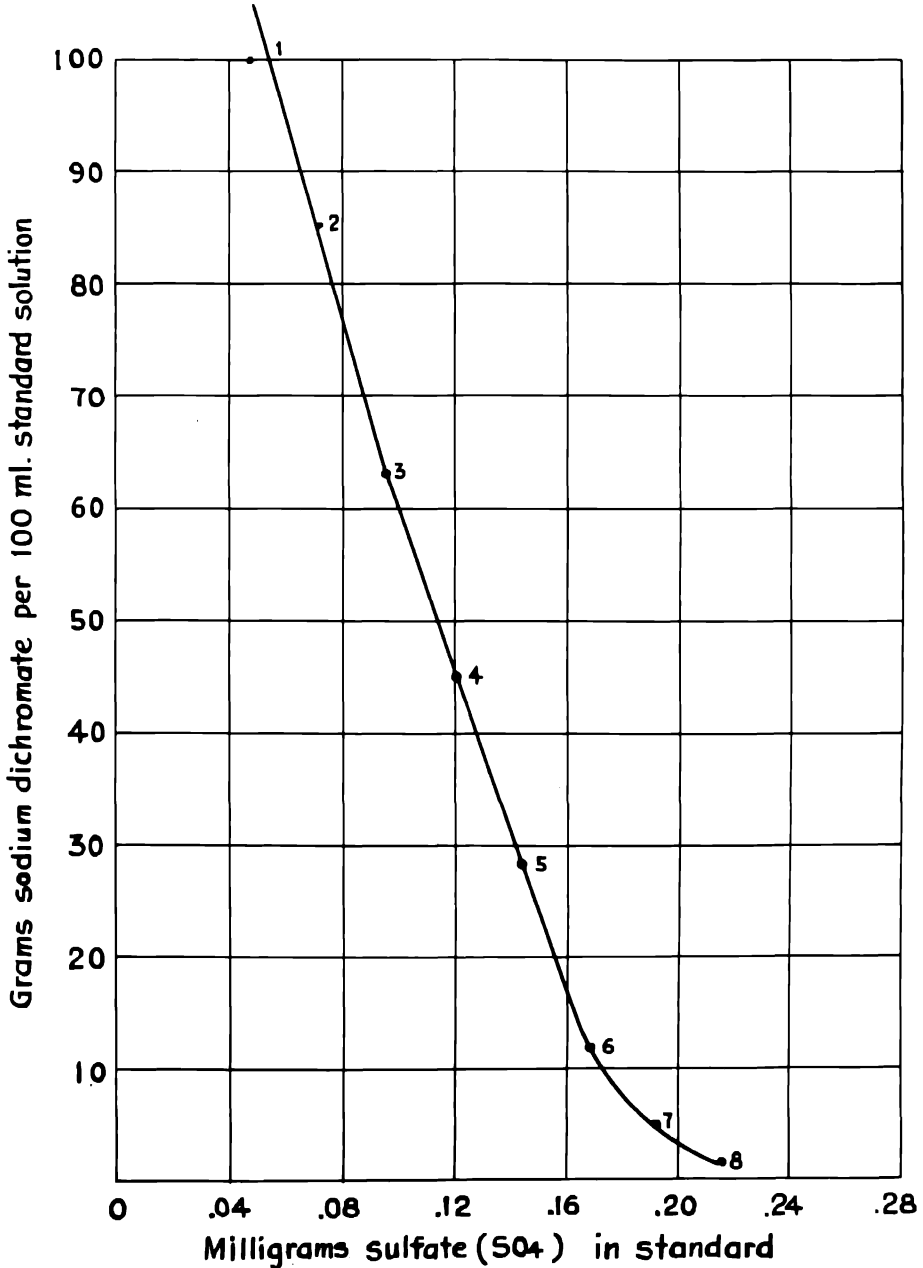


Fig. 1. Graph showing grams of sodium dichromate required to make 100 ml. of each sulfate standard and its sulfate equivalent in mg of  $\text{SO}_4$ .



The nearest approximations to the regularly developed test solutions were obtained by etching the outer surfaces of the vials in which the inorganic solutions were sealed. The etching was effected by dipping the stoppered vial, previously treated with cleaning solution, for five minutes in a proprietary etching compound ("Jack Frost").

*Preparation of Standards:* A concentrated aqueous solution of sodium dichromate is used as the base for the standards.

Solution A: Dissolve 250 grams C. P. sodium dichromate ( $\text{Na}_2\text{Cr}_2\text{O}_7 \cdot 2\text{H}_2\text{O}$ ) in distilled water and make up to a total volume of 250 ml. Filter.

Column 2 of Table I shows the quantity of this concentrate to use in making 100 ml. of each standard solution. Column 3 indicates the treatment given the vials in which the inorganic solutions are sealed. The sealing is effected by pouring molten paraffin into the vial filled within 20 mm. of the top with standard solution. A rubber stopper may be pushed into the opening in which case a little air space is left between the stopper and the paraffin.

TABLE I

Sulfate standard No.	ml. solution "A" per 100 ml. standard	Vial treatment	$\text{SO}_4$ equiv. of standard, in milligrams
1	100	Outer surface etched 5 minutes by "Jack Frost"	0.048
2	85		0.072
3	63		0.096
4	45		0.120
5	28		0.144
6	12		0.168
7	5	Unetched tube	0.192
8	1.5		0.216

Column 4 gives the sulfate equivalent in milligrams of  $\text{SO}_4$  of each standard. In Fig. 1, the sulfate content is plotted against the concentration of sodium dichromate in the standards. The smooth curve shows the regularity of the change of color to sulfate content and also serves as a confirmation of the figures experimentally obtained.

#### *Equipment Required:*

- 1 set sulfate color standards in box.
- 6 beakers, Pyrex glass, 100-ml. capacity.
- 6 funnels, glass, 90-mm. diameter.
- 1 volumetric flask, Exax, 10-ml. capacity.
- 1 volumetric flask, Exax, 25-ml. capacity.
- 1 vial block.
- 1 burette, Exax, 50-ml. capacity, for distilled water.
- 1 box Whatman No. 12, 15-cm. folded filter paper.
- 1 funnel rack, 10-hole.
- 1 phosphate illuminator.
- 2 pipettes, Mohr, 1-ml. capacity, graduated to 0.01 ml.
- 12 vials, shell, tall form, calibrated to 7.0 ml.
- 1 special pipette, 0.75-ml. capacity, with rubber bulb.

*Preparation of Special Equipment Required:* The tall-form shell vials are calibrated by filling from a 25-ml. burette to 7.0 ml. To prepare the special pipette, draw glass tubing of 7 mm. external diameter to a tip. It is calibrated by counting the number of drops equivalent to 1 ml., then drawing up 1 ml. from a 10-ml. calibrated graduate and letting out one-fourth of the number of drops determined. Scratch a mark at the 0.75-ml. point. Enlarge the upper end to accommodate a small rubber bulb.

*Reagents:*

*Sulfate Reagent No. 28, 0.01 N Barium Chloride:* Weigh out 1.2216 grams barium chloride ( $\text{BaCl}_2 \cdot 2\text{H}_2\text{O}$ ), wash into a liter volumetric flask and make up to the mark with distilled water. Preferably, make up a liter of tenth normal barium chloride solution and dilute 100 ml. to 1 liter.

*Sulfate Reagent No. 29, 0.1 Per Cent Aqueous Solution of Sodium Rhodizonate:* This reagent must be freshly prepared. It loses strength gradually and can be used for only a few hours. Portions of the salt are weighed out into small glass tubes. Take the amount needed and wash completely with distilled water into the size volumetric flask indicated. Make up to volume, stopper and shake until dissolved. Three-fourths ml. of the reagent added to 7.0 ml. of distilled water in the tall vial used for developing the color matches standard No. 8. If the preceding test shows a difference in the shade, the reagent must be discarded.

*Procedure:*

The volume of the samples tested and also of the various reagents used must be measured accurately. Since the volumes employed are exceedingly small, it is necessary to remove any liquid clinging to the outer surface of the pipettes before inserting the latter into the vials. A clean piece of filter paper is suggested for this purpose. Likewise, before the transfer, the tip of the pipette should be touched to the outer surface of the vial and after the transfer to the inner surface. This insures more accurate results.

1. Use fresh, untreated juice or juice to which has been added the preservative employed in rapid chemical methods.

2. The juice is mixed thoroughly and is filtered through Whatman No. 12, 15-cm. folded filter paper.

3. 0.20 ml. of the juice is transferred by means of a 1-ml. Mohr pipette, graduated to 0.01 ml., to the bottom of a tall vial (phosphate type).

4. 0.50 ml. of sulfate Reagent No. 28 is added by means of a pipette, similar to the one used above, to the bottom of the vial containing the juice; the contents are shaken for ten seconds.

5. Allow the vial to stand  $\frac{1}{2}$  minute.

6. Dilute with distilled water to the 7.0-ml. mark.

7. Stopper with the finger and mix by inverting three times.

8. Add 0.75 ml. of sulfate Reagent No. 29, using a specially calibrated pipette for the purpose.

9. Mix by inverting the vial three times. Let stand for 10 seconds, then compare with the aid of the phosphate illuminator against the sulfate standards.

10. If the developed solution is too red, use more juice and repeat Steps 3 through 9. If the solution is too yellow, use less juice and repeat Steps 3 through 9.

11. Use as many aliquots as possible, the colors of which fall within the range of the sulfate standards.

12. Record all the readings. Refer to Table II which gives the sulfate concentration in terms of parts per million for various aliquots used.

13. For the final result, take the average of the figures for the aliquots matching Standard No. 3 to Standard No. 8, inclusive.

### *Comparison of Results:*

The method has been applied to a number of representative cane juices secured in visits made to all of the plantations on Oahu. Both fresh and preserved juices were analyzed by the colorimetric method described and also by the regular gravimetric method. The results are shown in Table III.

TABLE II  
COLORIMETRIC DETERMINATION OF SULFATE IN CANE JUICE  
SULFATES ( $\text{SO}_4=$ ) IN PARTS PER MILLION

Standard No.	ml. sample used—													
	.05	.10	.15	.20	.25	.30	.35	.40	.45	.50	.60	.70	.80	1.00
1	960	480	320	240	192	160	137	120	107	96	80	69	60	48
2	1440	720	480	360	288	240	206	180	160	144	120	103	90	72
3	1920	960	640	480	384	320	274	240	213	192	160	137	120	96
4	2400	1200	800	600	480	400	343	300	266	240	200	171	150	120
5	2880	1440	960	720	576	480	411	360	320	288	240	206	180	144
6	3360	1680	1120	840	672	560	480	420	374	336	280	240	210	168
7	3840	1920	1280	960	768	640	548	480	426	384	320	274	240	192
8	4320	2160	1440	1080	864	720	617	540	480	432	360	309	270	216

TABLE III

Juice No.	Plantation	Variety	Treatment of sample	p.p.m. sulfate ( $\text{SO}_4=$ )—	
				Gravimetric	Colorimetric
1	Honolulu Pltn. Co.	31-2538	Fresh	1370	1450
2	Honolulu Pltn. Co.	31-2538	R.C.M. preservative	1350	1410
3	Oahu Sugar Co., Ltd.	H 109*	Fresh	868	886
4	Oahu Sugar Co., Ltd.	H 109*	Fresh	1026	1000
5	Oahu Sugar Co., Ltd.	28-3540	Preserved	576	592
6	Ewa Plantation Co.	H 109	Fresh	1112	1320
7	Ewa Plantation Co.	H 109	Preserved	1142	1200
8	Kahuku Plantation Co.	H 109	Fresh	1064	1220
9	Kahuku Plantation Co.	H 109	Preserved	1066	1160
10	Waianae Company	H 109	Fresh	1020	1160
11	Waianae Company	H 109	Preserved	1014	1130
12	Waialua Agr. Co., Ltd.	H 109	Fresh	822	910
13	Waialua Agr. Co., Ltd.	H 109	Preserved	834	906
14	Waialua Agr. Co., Ltd.	H 109	Fresh	710	735
15	Waialua Agr. Co., Ltd.	H 109	Preserved	756	705

\* Tops included.

It will be noted that although the colorimetric results vary somewhat from the gravimetric figures, the variation is within the limits of the change from one standard to the next.

## SUMMARY

A colorimetric method for the determination of sulfate in cane juice is described, including complete details of technic, instructions for making permanent inorganic standards and comparisons with gravimetric results. The method is rapid and can be carried out by trained workers.

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## The Third Study of Water and Cane Ripening

By CONSTANCE E. HARTT

The first and second reports of an investigation of the importance of water in the ripening of cane were published in 1934 (1) and 1936 (4). The plants used in the third study were the ratoons of those used in the second study. A brief account of the results obtained in the third study has already been rendered (6). All of the experiments illustrate the importance of water in increasing the amount of sugar produced by the leaves, in facilitating the transport of sugar from the leaves to the stems, and in aiding the expression of sugar in the juice.

### METHODS

Sugar cane plants of the variety H 109, planted in pots of good soil at the Experiment Station on October 8, 1934, were ratooned on November 30, 1935, and given complete fertilization. The plants were uniformly watered twice daily for eleven months. Their growth was excellent and uniform, showing no residual effect of treatment.

On September 30, 1936, the following five series were inaugurated, with 14 pots per series:

1. Dark wet
2. Dark dry
3. Light wet
4. Light dry
5. Outdoor control

The plants in each series were taken from all parts of the plot. No attempt was made to use the root systems and ratoons therefrom of a given series in the second experiment for the same series in the third experiment.

On Wednesday, September 30, the plants of series 1-4 were placed in the greenhouse. The plants of all series were watered twice daily until Thursday noon, October 8, after which time the plants of series 2 and 4 received no more water.

Growth measurements of all plants were made twice daily from October 4 to 10 inclusive, and a final measurement on October 12. The measurements were made by Frederick F. Hébert and William O. Smith. The measurement used was the distance to the highest emerged dewlap from a constant base reference mark. Two representative stalks in each pot in each series were measured at 8 a. m. and 2 p. m. The results for each series were averaged and plotted. The graphs were used as a basis for deciding when to continue the experiment, since it has been shown by Wadsworth (9, 10) that the growth curve for sugar cane flattens out when the soil in which the cane is grown reaches the wilting point. Such a flattening of the curves following the withdrawal of water is shown in Figs. 2 and 4. The growth of the plants of series 2 and 4 had ceased by October 10, and their leaves were rolled and dried at the tips, whereas the plants of series 1, 3 and 5 continued to grow and their leaves continued to be turgid.

The plants of series 1-4 were placed in the darkened assembly room on October 10, being moved from 2-4:15 p. m. The plants of series 3 and 4 were returned to the greenhouse on October 11, being moved from 6:30-7:45 p. m. These plants received light from 6:15 a. m. until 1 p. m. October 12, when they were sampled. The plants of series 1, 3 and 5 were watered twice daily throughout the experiment.

The plants of series 1 and 2 were sampled at 8 a. m., October 12, while still in the darkened assembly room, with the aid of flashlights. The plants of series 3, 4 and 5 were sampled at 1 p. m. the same day.

In sampling, two stalks were taken from each pot, care being exercised not to select the stalks reserved for growth measurements. Counting the leaf with the highest emerged dewlap as leaf number 1, leaves number 1 and 2 were taken. The entire green-leaf cane was used. Seven stalks of dry-leaf cane from each series were taken at random, cleaned, split lengthwise, and half used in sampling. The other complete stalks were used for juice analyses without being cleaned, the juice being expressed in a Cuba mill.

The order of sampling was as follows: first the blades, then the sheaths, then the green-leaf cane, and finally the dry-leaf cane. The series were sampled in numerical order. Samples were taken for the estimation of moisture, sugars, and enzymes.

Soil samples for the determination of moisture content were taken from series 1-4 on October 12 and 13. The plants were then discarded.

Determinations were made of simple sugars and sucrose by methods described previously (5). Determinations of the activities of the enzymes invertase, amylase, dextrinase, and maltase were made by methods already outlined (4). An improvement in technique was used in the determinations of invertase and maltase, involving the use of double controls. These double controls wipe out apparent but unreal differences in activity which are caused by variations in sugar content in the plant material used in the determination of enzyme activity.

The plan of the experiment included determinations of starch and total polysaccharides. The plant material for these determinations was ground to 100 mesh in a ball mill and stored in aluminum tins until analyzed. Unfortunately the finely ground material adsorbed aluminum from the tins, which inhibited the activity of the taka-diestase used in the analysis. Several samples were lost due to breakage of the glass bottles of the ball mill. For these reasons incomplete and unsatisfactory results were obtained for starch and polysaccharides. Therefore, we are unable to calculate the results upon the usual residual dry-weight basis, because for that method of calculation one must know the percentage of polysaccharides. The usual residual dry weight is calculated by subtracting the sum of the total sugars plus polysaccharides from the dry weight. Instead, we are reporting results upon a modified residual dry-weight basis, in which the total sugars alone are subtracted from the dry weight. The results by all three methods of calculation show the same tendencies almost without exception. The differences are intensified when calculated on the modified residual dry-weight basis. For this reason, and also because it is considered the most accurate method of calculation, the discussion of results will be based upon the modified residual dry-weight method of calculation. The results were also calculated on the water basis, i. e., grams sugar per 100 grams water, but are not so expressed here because they showed no essential dif-

ference from the other methods of calculation. Because both the sugar and the moisture percentages varied, the results on the water basis depend upon two variables and are therefore not as accurate a representation of the actual changes in sugar content as the results by the modified residual dry-weight method.

## RESULTS

The measurements made of the highest emerged dewlap of two stalks per pot were averaged for each series and are plotted in Figs. 1-5. These measurements were made for four days before water was withheld, for two days after water was withheld, and on the final day of the experiment. The straight line curves in Figs. 1, 3 and 5 are indicative of uninterrupted growth in the plants supplied with water throughout the experiment. Figs. 2 and 4 show that elongation ceased one day after watering was discontinued and there was no further increase in length during the experiment. The growth curves therefore indicate that at the time of the experiment, October 12, the day the plants were supposed to conduct photosynthesis, the soils of series 2 and 4 were at or below the wilting point and the soils of series 1, 3 and 5 were well above that point.

Conclusive evidence to that effect is afforded by the actual determinations of the soil moisture content. Wadsworth (11) reported the following results for the average moisture content of the soil in series 1-4:

Series 1:	36.2%
2:	21.0%
3:	26.7%
4:	20.2%

Assuming that the permanent wilting per cent is 25.7, series 1 and 3 were well above and series 2 and 4 were well below the wilting point. The result for series 3 is below that for series 1 because some of the pots in series 3 were not sampled until October 13, whereas all the other pots of all the other series were sampled on October 12, the day of the experiment.

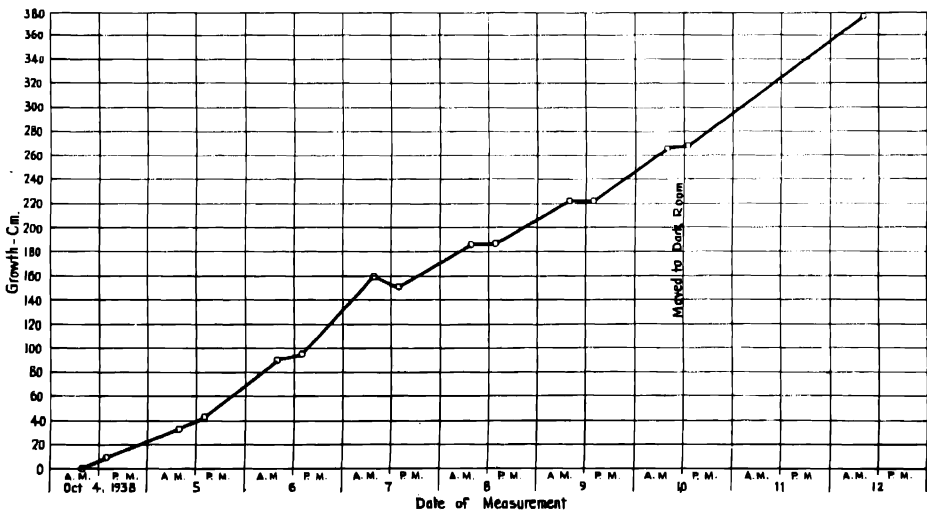


Fig. 1. Increase in length of series 1.



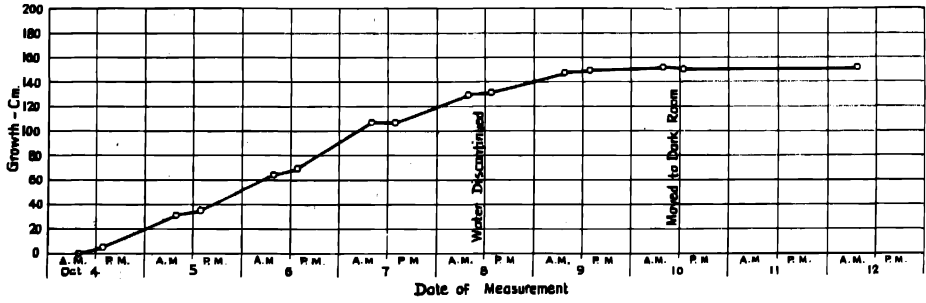


Fig. 2. Increase in length of series 2.

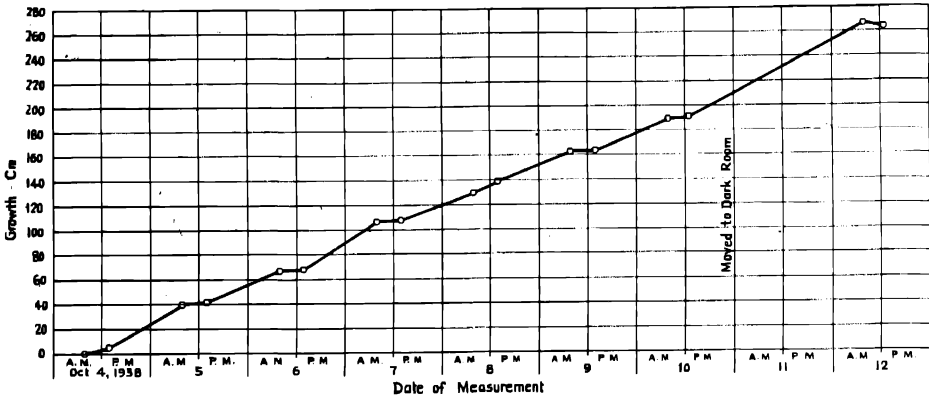


Fig. 3. Increase in length of series 3.

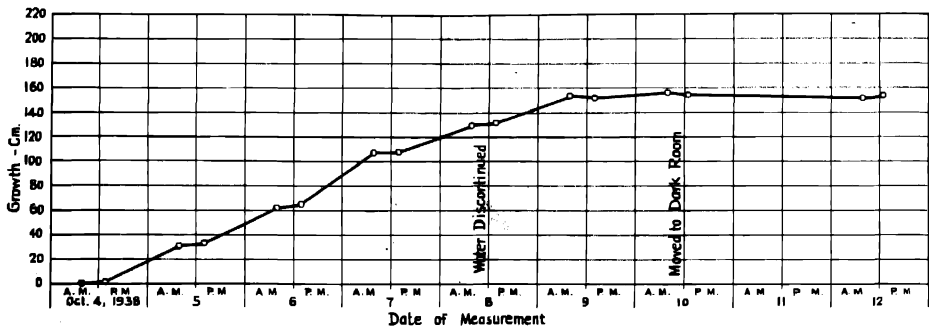


Fig. 4. Increase in length of series 4.

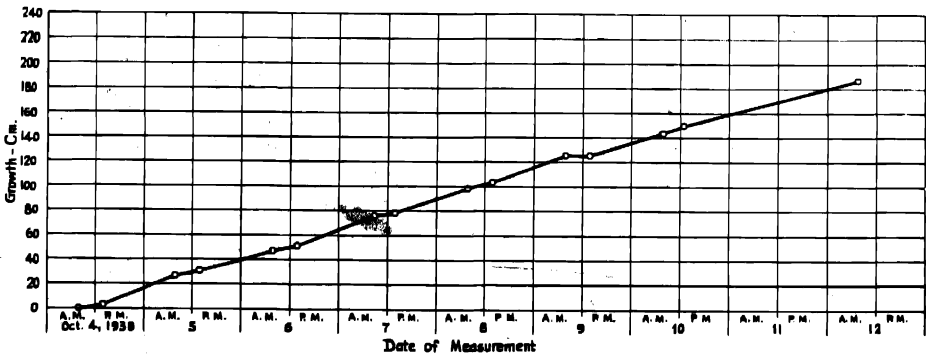


Fig. 5. Increase in length of series 5.



Fig. 6. Appearance of the plants which were watered twice daily. Photographed October 10, 1936.



Fig. 7. Appearance of the plants which had received no water for  $1\frac{1}{2}$  days.  
Photographed October 10, 1936.



Fig. 8. Appearance of the plants which had received no water for  $1\frac{1}{2}$  days.  
Photographed October 10, 1936.

The data obtained from the growth measurements and soil moisture determinations prove conclusively that we were contrasting plants in soil well above the wilting point with plants in soil at or below the wilting point, which was the object of the experiment.

The plants were photographed on October 10, 1½ days after water was withheld. The plants which were watered twice daily are illustrated in Fig. 6; those which had been deprived of water for 1½ days are illustrated in Figs. 7 and 8, and the rolling of the leaves is particularly conspicuous at the tops of the plants.

TABLE I  
JUICE ANALYSES OF THE DRY-LEAF CANE

Series	Brix	Pol	Sucrose	Apparent purity	Gravity purity	Glucose	Q.R.
1. Dark wet .....	18.25	16.34	16.57	89.53	90.79	0.68	8.06
2. Dark dry .....	17.19	14.63	14.95	85.11	86.97	1.08	9.29
3. Light wet .....	17.68	15.38	15.64	86.99	88.46	0.96	8.71
4. Light dry .....	17.40	15.15	15.45	87.07	88.79	0.96	8.84
5. Outdoor control .....	18.98	17.25	17.50	90.89	92.20	0.56	7.56

The results of the juice analyses, which were conducted by the Sugar Technology department, are recorded in Table I, which shows that the plants of series 5 (outdoor control) had the most sucrose, the least glucose, and the best quality ratio. The plants supplied with water contained more sucrose and had a better quality ratio than the plants deprived of water, in both the dark and the light.

If the results obtained with the plants in the dark are subtracted from the results obtained with the plants in the light, the increase or decrease during the time of exposure to light is obtained. This has been done and the results are presented in Table II. Table II shows that the dry-leaf cane of the plants supplied with water deteriorated during the day in every quality tested, whereas the dry-leaf cane of the plants deprived of water improved in every category. Notwithstanding these facts, the juice of the plants supplied with water was superior to the juice of the plants deprived of water, both in the dark and in the light.

TABLE II  
INCREASE OR DECREASE DURING EXPOSURE TO LIGHT FOR 7 HOURS,  
DRY-LEAF CANE

Series	Brix	Pol	Sucrose	Apparent purity	Gravity purity	Glucose	Q.R.
Supplied with water.....	—57	—96	—93	—2.54	—2.33	+28	+65
Deprived of water.....	+21	+52	+50	+1.96	+1.82	—12	—35

The results of the moisture determinations are reported in Table III, which shows that the plants supplied with water had higher moisture percentages than the plants deprived of water in all organs tested, with the exception of the green-leaf cane in the light. The blades and sheaths of the plants in the dark had higher percentages of moisture than those in the light. This difference was not found in the cane, however, in which for the most part the plants in the light had the higher percentages of moisture.

TABLE III  
MOISTURE DETERMINATIONS

Series	Blades	Sheaths	Green-leaf cane	Dry-leaf cane
Percentages expressed on wet-weight basis				
1. Dark wet .....	69.09±0.007	79.81±0.281	84.50±0.076	72.82±0.062
2. Dark dry .....	65.95±0.277	75.47±0.005	80.64±0.019	71.93±0.038
3. Light wet .....	64.92±0.005	76.21±0.000	81.41±0.167	74.07±0.043
4. Light dry .....	63.71±0.176	71.39±0.100	82.03±0.024	73.47
5. Outdoor control .....	64.45±0.029	74.82±0.043	81.53±0.007	71.70±0.029
Percentages expressed on dry-weight basis				
1. Dark wet .....	223.6 ±0.048	394.3 ±7.680	545.4 ±3.244	268.0 ±0.859
2. Dark dry .....	193.5 ±1.717	307.7 ±0.048	416.6 ±0.477	256.3 ±0.525
3. Light wet .....	185.0 ±0.024	320.3 ±0.000	438.2 ±4.865	285.7 ±0.668
4. Light dry .....	175.6 ±1.336	246.4 ±2.719	456.7 ±0.811	276.9
5. Outdoor control .....	181.3 ±0.238	297.3 ±0.715	441.6 ±0.143	253.4 ±0.382

The results of the determinations of simple sugars are presented in Table IV and of sucrose in Table V. The increases and decreases in simple sugars and sucrose during exposure to light for seven hours, upon the modified residual dry-weight basis, are recorded in Table VI.

TABLE IV  
SIMPLE SUGARS: PERCENTAGES EXPRESSED ON THE WET-WEIGHT BASIS, THE DRY-WEIGHT BASIS, AND THE MODIFIED RESIDUAL DRY-WEIGHT BASIS

Series	Wet weight	Dry weight	Modified residual dry weight
Blades:			
1. Dark wet .....	0.366±0.003	1.184±0.009	1.223±0.010
2. Dark dry .....	0.435±0.006	1.279±0.017	1.317±0.018
3. Light wet .....	0.303±0.001	0.866±0.004	0.914±0.004
4. Light dry .....	0.474±0.000	1.307±0.001	1.350±0.001
5. Outdoor control .....	0.349±0.001	0.984±0.003	1.028±0.003
Sheaths:			
1. Dark wet .....	1.669±0.001	8.268±0.005	9.537±0.006
2. Dark dry .....	1.552±0.010	6.327±0.042	7.076±0.048
3. Light wet .....	1.360	5.548	6.237
4. Light dry .....	1.449±0.011	5.066±0.039	5.564±0.043
5. Outdoor control .....	1.356±0.000	5.388±0.002	6.164±0.002
Green-leaf cane:			
1. Dark wet .....	2.632	16.982	28.488
2. Dark dry .....	2.571±0.000	13.286±0.004	22.507±0.006
3. Light wet .....	2.606±0.003	13.887±0.050	24.085±0.085
4. Light dry .....	3.047±0.000	16.957±0.002	28.459±0.004
5. Outdoor control .....	2.740±0.020	14.856±0.120	24.374±0.197
Dry-leaf cane:			
1. Dark wet .....	0.255±0.000	0.938±0.000	1.825±0.001
2. Dark dry .....	0.419±0.004	1.495±0.016	3.145±0.033
3. Light wet .....	0.687±0.001	2.651±0.006	5.165±0.012
4. Light dry .....	0.725±0.000	2.735±0.001	5.647±0.003
5. Outdoor control .....	0.417±0.002	1.476±0.007	2.830±0.013

The results of the determinations of the activity of invertase are presented in Table VII. There was no evidence that the activity of invertase was affected by treatment.

TABLE V

CANE SUGAR: PERCENTAGES EXPRESSED ON THE WET-WEIGHT BASIS, THE DRY-WEIGHT BASIS, AND THE MODIFIED RESIDUAL DRY-WEIGHT BASIS

Series	Wet weight	Dry weight	Modified residual dry weight
Blades:			
1. Dark wet .....	0.603±0.007	1.952±0.023	1.915±0.023
2. Dark dry .....	0.565±0.008	1.659±0.026	1.624±0.025
3. Light wet .....	1.559±0.004	4.445±0.012	4.460±0.012
4. Light dry .....	0.694±0.011	1.914±0.031	1.879±0.030
5. Outdoor control .....	1.169±0.001	3.126±0.002	3.436±0.002
Sheaths:			
1. Dark wet .....	1.016±0.001	5.032±0.005	5.514±0.006
2. Dark dry .....	1.041±0.001	4.245±0.003	4.507±0.002
3. Light wet .....	1.378	5.616	6.314
4. Light dry .....	1.112±0.009	3.887±0.033	4.055±0.035
5. Outdoor control .....	1.814±0.024	7.202±0.094	7.828±0.102
Green-leaf cane:			
1. Dark wet .....	3.625	23.407	37.304
2. Dark dry .....	5.359±0.000	27.681±0.003	44.545±0.005
3. Light wet .....	5.259	28.563	47.062
4. Light dry .....	4.215±0.014	23.457±0.076	37.398±0.122
5. Outdoor control .....	4.470±0.016	24.199±0.087	37.723±0.136
Dry-leaf cane:			
1. Dark wet .....	12.955±0.023	47.666±0.085	88.105±0.158
2. Dark dry .....	14.317	51.003	101.932
3. Light wet .....	11.932±0.029	46.017±0.111	85.163±0.205
4. Light dry .....	12.953±0.074	48.824±0.278	102.204±0.296
5. Outdoor control .....	13.627±0.013	48.152±0.045	90.814±0.084

TABLE VI

INCREASE OR DECREASE IN SUGARS DURING EXPOSURE TO LIGHT FOR 7 HOURS, EXPRESSED UPON THE MODIFIED RESIDUAL DRY-WEIGHT BASIS

Series	Blades	Sheaths Simple Sugars	Green-leaf cane	Dry-leaf cane
Supplied with water.....	— .309	— 3.300	— 4.403	+ 3.340
Deprived of water.....	+ .033	— 1.512	+ 5.952	+ 2.502
		Sucrose		
Supplied with water.....	+ 2.545	+ .800	+ 9.758	— 2.942
Deprived of water.....	+ .255	— .452	— 7.147	+ .272

TABLE VII

INVERTASE ACTIVITY EXPRESSED IN C.C. N/20 KMnO<sub>4</sub>

Series	Blades	Sheaths	Green-leaf cane	Dry-leaf cane
	Activity unbuffered			
1. Dark wet .....	8.36	18.36	1.30	0.00
2. Dark dry .....	8.40	15.78	3.23	0.00
3. Light wet .....	8.00	13.83	0.10	0.30
4. Light dry .....	7.86	15.17	2.65	0.00
5. Outdoor control .....	8.55	13.22	0.00	0.10
	Activity at pH 4.5			
1. Dark wet .....	13.75	17.64	1.24	0.00
2. Dark dry .....	13.25	19.48	3.97	0.00
3. Light wet .....	12.29	18.59	0.00	0.00
4. Light dry .....	14.18	18.05	1.75	0.62
5. Outdoor control .....	12.31	18.25	0.00	0.00

The activity of amylase was tested unbuffered and at a series of reactions ranging from pH 4.5-5.8. No activity was demonstrated in the green-leaf cane or the dry-leaf cane. The optimum reaction for amylase in blades was found to be pH 5.2, at which reaction the activity in decreasing order was: 5, 1 and 4, 2 and 3. In sheaths, the activity of amylase was very weak; there was no effect of hydrogen ion concentration and no effect of treatment.

The activity of dextrinase was determined unbuffered and at a series of reactions ranging from pH 4.6 to 8.0. No dextrinase was detected in the green-leaf cane or the dry-leaf cane. The optimum reaction for dextrinase in blades was pH 5.4-5.8; series 1 and 5 were a little more active than the other series, but they were all very weak. In sheaths, dextrinase was very weak; there was no effect of hydrogen ion concentration and no effect of treatment.

Maltase activity was very weak or absent in all four organs.

## DISCUSSION

### *Blades:*

The blades of the plants supplied with water, as well as those deprived of water, lost water during the seven-hour period of exposure to light. This loss was only to be expected, because the moisture content of sugar cane blades grown under natural conditions reaches its lowest point in the early afternoon (2). The blades of the plants supplied with water had higher percentages of moisture than those of the plants deprived of water.

The recorded changes in simple sugars were probably insignificant.

The increase in sucrose in the blades of the plants supplied with water was ten times that in the blades of the plants deprived of water, which was barely significant.

The very small but significant increase in simple sugars and sucrose in the blades deprived of water indicated that very little photosynthesis took place in them.

These results show definitely that sugar cane requires a plentiful supply of water for the production of cane sugar in the leaves.

### *Sheaths:*

The sheaths of both treatments lost water during the day, as do sheaths of plants grown in the open (3). The sheaths of the plants supplied with water had higher moisture percentages than those of the plants deprived of water.

The sheaths of both series lost water and lost simple sugars during the day.

The percentage of sucrose increased a little in the sheaths of the plants supplied with water, and decreased a little in the sheaths of the other plants.

### *Green-leaf cane:*

The green-leaf cane of the plants supplied with water decreased in percentage of moisture during the day, thus resembling the blades and the sheaths. The green-leaf cane of the plants deprived of water, however, gained a little in percentage of moisture, the explanation of which is not known. A possible source of this small increase in water is the release of water bound to the complex colloids in the lower leaves attached to the green-leaf cane, many of which died during the experiment. Whatever the source, it was internal rather than external, for it is



certain that no water was supplied to the plants deprived of water during the course of the experiment.

The percentage of simple sugars decreased in the green-leaf cane of the plants supplied with water but increased in the plants deprived of water.

The percentage of sucrose increased in the plants supplied with water but decreased in the plants deprived of water. The chief cause of this difference was probably the greater supply and more rapid movement of sucrose from the blades of the plants supplied with water than from those of the plants deprived of water. Another cause was the formation of sucrose from glucose, since that process is known to take place in green-leaf cane (7, 8). The green-leaf cane of the plants supplied with water decreased in moisture content, decreased in simple sugars, and increased in sucrose, indicating that simple sugars condensed to sucrose. The green-leaf cane of the plants deprived of water increased in moisture, increased in simple sugars, and decreased in sucrose, indicating that sucrose was inverted to simple sugars. Therefore, small changes in moisture content of the green-leaf cane affect the equilibrium between the simple sugars and sucrose.

#### *Dry-leaf cane:*

The dry-leaf cane of both series gained a little moisture during the day. The percentage of simple sugars increased in both series. The percentage of sucrose decreased in the plants supplied with water, but did not change significantly.

Comparison will now be made of the results obtained on the tissue basis with those obtained by juice analysis. On the tissue basis, the dry-leaf cane of the plants deprived of water contained *more* sucrose than that of the plants supplied with water. On the juice basis, however, the dry-leaf cane of the plants deprived of water contained *less* sucrose than that of the plants supplied with water. Evidently, not all of the sucrose in the cane of the plants deprived of water was extracted by the Cuba mill. These results agree with those of the second experiment (4) in indicating that a plentiful supply of water aids in the expression of sugar in the juice.

The sucrose content of the dry-leaf cane of the plants deprived of water was not only greater than that of the plants supplied with water, but also greater than that of the outdoor control. It is evident that the increase in sucrose in the dry-leaf cane of the plants deprived of water did not occur during the day of the experiment (i. e., October 12), but rather took place during the 4-day period when water was withheld.

The results presented in Tables I-V show that the dry-leaf cane was not static. The time of day, water supply, and removal of the plants from the outside to the greenhouse are some of the factors which have been found to affect the juices. The Brix, polarization, sucrose, purity, glucose and quality ratio of the juices were affected, as well as the percentages of moisture, simple sugars, and sucrose in the tissue of the dry-leaf cane. Wadsworth (10) found a diurnal change in Brix in the dry-leaf cane of plants grown in a hot, dry environment in irrigated areas, and considered that the changes in Brix were due to diurnal changes in moisture content rather than to changes in sugar content. The results herein reported indicate that these differences are not caused solely by changes in water content but are actual differences in percentage of sugar on the dry-weight basis. Sucrose once laid down in the dry-leaf cane does not remain unchanged; it may be added

to or subtracted from. However, the fluctuations in sucrose content of the dry-leaf cane are less than those of the green-leaf cane.

#### *Enzyme activity:*

In the second study of water and cane ripening (4) it was found that the enzymes in the blades were sensitive to differences in moisture and light; the enzymes in the sheaths were less sensitive than those in the blades; maltase was the only enzyme affected in the green-leaf cane; and in the dry-leaf cane the differences were insignificant.

In the experiment reported now, there was no evidence that the activities of invertase or maltase were affected by treatment. Differences in the activity of amylase and dextrinase were small and were not consistent with those obtained in the second experiment. In short, the results of the determinations of enzyme activity presented now do not support the results obtained in the second experiment.

One cause of the differences in the enzyme results obtained in the two experiments lies in the difference in technique mentioned in "Methods." Because the double controls were used in this experiment but not in the second experiment, it is felt that the results of this experiment are more reliable than those of the second experiment. Therefore, we have as yet no evidence of differences in enzyme activity resulting from differences in moisture content due to supplying or withholding water for four days. This does not mean that drying out has no effect upon enzyme action. The leaves taken for analysis were numbers 1 and 2 (see Methods), and were not the leaves severely affected by lack of water, which were the lower leaves. In fact, the moisture percentage of the blades of the plants deprived of water did not vary greatly from that of the blades of the outdoor controls, being a little more in the dark and a little less in the light. Another study by a different method is now under way, one object of which is to find the effect of different supplies of water upon the activity of enzymes in the sugar cane plant.

#### *Which is the better plant?*

The plants supplied with water were definitely better producers of cane sugar than the plants deprived of water, for the blades of the former made ten times as much sucrose as the blades of the latter, the green-leaf cane of the former gained a large amount of sucrose while that of the latter lost sucrose, and the juice extracted from the dry-leaf cane of the former had a better quality ratio than that from the latter. It is true that on the tissue basis the dry-leaf cane of the plants deprived of water had the higher percentage of sucrose; but if the sucrose is left in the pulp when the juice is extracted in the mill, it is a loss rather than a gain. Perhaps the extraction of sugar from ripened cane requires a method different from the extraction of sugar from cane of high water content. Whether or not the addition of water to the pulp from ripened cane would result in a gain in extraction of sucrose sufficient to be of economic importance is problematical.

#### SUMMARY

The third study of water and cane ripening confirms the conclusions of the first two studies: a plentiful supply of water is essential for the formation of

sucrose in the blades, for its transport to the stem, and for the expression of sugar in the juice.

Plants in soil at or below the wilting point may carry on the process of photosynthesis, but the sugar made is very much less than that made by plants adequately supplied with water. What sugar is made is stored rather than used, the result being a higher percentage of cane sugar in the dry-leaf cane of the plants deprived of water than in that of the other plants. This gain in sucrose is of physiological interest but probably not of economic importance because it is not all extracted in the juice.

Small changes in moisture content of the green-leaf cane affect the equilibrium between the simple sugars and sucrose.

The sugar content of the dry-leaf cane is not static but is affected by such factors as the time of day, the water supply, and the removal of plants from the outside to the greenhouse.

#### ACKNOWLEDGMENTS

The writer is indebted to many people who have helped in this investigation. Ada Forbes was of constant assistance during the entire course of the study. Ingrid Larsen helped with many of the analyses. Dr. A. J. Mangelsdorf furnished help for the care of the plants. Professor H. A. Wadsworth collaborated in the investigation, and with his assistants made soil moisture determinations. Frederick F. Hébert and William O. Smith took all the growth measurements. The juice analyses were performed by the Sugar Technology department. Wm. Sa Ning moved the plants and darkened the assembly room. Walter Ah You assisted in many ways.

The writer desires particularly to thank Dr. H. L. Lyon for all his help and advice in carrying on this investigation.

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## Sugar Prices

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96° CENTRIFUGALS FOR THE PERIOD  
JANUARY 3, 1939 TO MARCH 15, 1939

	Date	Per pound	Per ton	Remarks
Jan.	3, 1939..	2.80¢	\$56.00	Philippines.
"	5.....	2.825	56.50	Philippines, 2.80; Cubas, 2.85.
"	10.....	2.75	55.00	Puerto Ricos.
"	18.....	2.77	55.40	Philippines, Puerto Ricos.
"	20.....	2.82	56.40	Cubas.
"	24.....	2.805	56.10	Puerto Ricos, 2.80 and 2.81.
"	25.....	2.80	56.00	Puerto Ricos.
"	26.....	2.78	55.60	Puerto Ricos.
Feb.	2.....	2.775	55.50	Puerto Ricos, 2.77; Philippines, 2.78.
"	3.....	2.75	55.00	Puerto Ricos.
"	10.....	2.76	55.20	Philippines.
"	11.....	2.755	55.10	Philippines, 2.76; Puerto Ricos, 2.75.
"	14.....	2.75	55.00	Philippines.
"	15.....	2.76	55.20	Philippines.
"	20.....	2.755	55.10	Philippines, 2.75; Puerto Ricos, 2.75 and 2.76.
"	21.....	2.80	56.00	Cubas.
"	25.....	2.78	55.60	Philippines.
"	27.....	2.80	56.00	Puerto Ricos, Philippines.
Mar.	7.....	2.77	55.40	Philippines.
"	8.....	2.78	55.60	Philippines.
"	10.....	2.77	55.40	Cubas, 2.78; Philippines, 2.77; Puerto Ricos, 2.76.
"	13.....	2.78	55.60	Puerto Ricos.
"	15.....	2.80	56.00	Puerto Ricos, Philippines.

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# THE HAWAIIAN PLANTERS' RECORD

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THIRD QUARTER 1939

No. 3

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## **In This Issue:**

### *Nitrogen in the Cane Leaf:*

A method is described which is, at present, under development as a means of estimating the nitrogen requirement of sugar cane. The procedure of study, first proposed and inaugurated by H. P. Agee, includes an R.C.M. analysis of specific cane leaf tissue obtained by a ticket punch. A relationship appears to exist between the nitrogen content of the leaf tissue taken from a definite region on the cane stalk, and growth of the plant. The results of this study have contributed to improvement of experimental technic which, it is hoped, may eventually lead to the establishment of a practical control in nitrogen fertilization of sugar cane. The method is based upon making periodic ticket-punch collections of leaf specimens in the field, obtaining their total nitrogen content by R.C.M. analysis and estimating the needs of the crop for nitrogen by interpreting the data with respect to the *sufficiency* of the nutrient at different ages of growth. Potential application in practice of the system is discussed, aided by data presented graphically.

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### *Dead Cane at Harvest:*

The factors causing cane to die prior to harvest are at times obvious but there are many instances where the causal factor or factors are not understood. In this paper an attempt has been made to list and briefly discuss the more common factors responsible for dead cane at harvest.

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### *The Effects of Oven Drying and Air Drying on the Available Nitrogen Content of Soils:*

A resume is presented of a study made to determine the extent and character of changes taking place in the concentration of *available* soil nitrogen as a result of natural or artificial drying of the soil preparatory to chemical analysis.

A rapid chemical method is described for the determination of *total available* nitrogen in the freshly collected, moist soil specimen.

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*Sunlight-Nitrogen Relationships:*

Interesting evidence is presented of the separate effects upon cane growth and quality from variations in periods of exposure to direct sunlight and from different amounts of nitrogen, but no significant interaction between these two factors was brought out in this first of a series of skirmish tests concerned with these issues.

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## Nitrogen in the Cane Leaf

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By Q. H. YUEN and FRANCIS E. HANCE

*A study of some relationships existing between the total nitrogen of a specific region of the cane plant, considering associated meteorological influences, and with related factors probably affecting crop growth, fertilizer requirements, maturing and, as ripening sets in, with expected quality of crusher juice.*

*The determination of the "nitrogen index" of a field of cane by an organized collection and R.C.M. analysis of ticket-punch leaf specimens secured at monthly intervals currently with the development of the crop.*

*An attempted development of a plantation field-scale nitrogen fertilizer control (potash and phosphate assumed not limiting factors).*

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Cane tonnage in sugar production in Hawaii has increased considerably during the past twenty years. However, the number of tons of cane required to produce a ton of sugar has also increased and hence economy of production has fallen off. Employment of new varieties, improved cultural practices and liberal use of fertilizers have, of course, been responsible to a large extent for advances made in increased tonnage and at times for improvement in yield.

From the standpoint of an efficient fertilizer practice, nitrogen applied in excess of crop requirement has been considered by many as having favored cane tonnage at the expense of cane quality. The effect of nitrogen on the sucrose content of sugar cane grown under field conditions was made a subject of thorough investigation by Alexander (2), Verret (16) and Borden (3). In general, their conclusions coincide in that increasing increments of nitrogen applied above a certain minimum optimum have a depressing effect upon quality of juice.

The subject of control in nitrogen fertilization is, therefore, one of importance both from the standpoint of cultural practice and from related economic aspects.

Control of crop fertilization may be based on field experiment, laboratory cultural experiment, soil analysis, plant analysis, or by rule of thumb—the expedient commonly resorted to in nitrogen fertilization. However, limitations are inherent in all such procedures, but a degree of reliability may be realized by combining a number of them or by a correlation study of the whole series of data. Another modification of crop fertilization control embraces an interrelated soil and plant material analysis. Such a method was proposed in a paper by Lundegårdh (11).

A modicum of success in the control of crop fertilization by soil analysis has been achieved for phosphate and potash on Hawaiian sugar plantations. In the case of nitrogen, only partial control can be exercised by this means due to the characteristic behavior of nitrogen in the soil under Hawaiian conditions. This subject has been discussed by Yuen and Borden (17) in a recent publication. They point out that a determination of the *available* nitrogen in the soil will only indicate soil fertility for this nutrient at the moment of collection and that nitrogen applied to a



growing crop will not remain in the soil for any appreciable period of time due to rapid absorption by the crop—an established fact—and due also, it appears, to shifty availability occasioned by chemical reaction, microbiological activity, or loss by leaching. Dean (8) and his associates have shown that ammonia and nitrate nitrogen, available to plant life, are produced under field conditions from the organic nitrogen reserves of the soil. Pending the completion of Dean's researches and the development of R.C.M. procedures for evaluating these factors, chemical analysis of soils for the estimation of the crop requirement or its available supply of nitrogen, we believe, can only be partially sufficient. Incidentally, the researches of Dean and his coworkers have resulted in the development of a method to determine the quantity of "mineralizable nitrogen," as they describe it, in a soil by subjecting the soil to controlled incubation under laboratory conditions and thereafter determining the amount of available nitrogen formed after a stated period.

If immediate control in nitrogen fertilization is to be attempted in the cane field, it appears that soil analysis should be supplemented by analysis of the plant itself, provided reliability of the procedure can be secured. Another factor which enters into a consideration of nitrogen nutrition is the effect of weather on the behavior of this nutrient in the plant and in the soil which supports it. A discussion of the relationship of climate to sugar production in Hawaii has been published by Das (4, 5). From these and later studies (6) the inference may be gathered that climatic conditions of temperature and sunlight play important roles in the growth processes of the plant, which in turn may affect the nitrogen requirement of the crop. With favorable weather conditions, large applications of nitrogen may be utilized by cane, apparently, and heavier tonnages secured without undue detrimental effects upon the quality of crusher juice. During periods or cycles of unfavorable growing weather, less nitrogen may be supplied to the crop if the detrimental factor of luxury consumption and consequent effects upon ripening and juice quality are to be avoided. The nitrogen requirement of sugar cane, therefore, appears to be one that is variable. Any control of this requirement that would prove satisfactory should be one that reflects or indicates such variability. Attention was therefore directed to the cane plant itself. Pursuit of this objective has engaged the attention of Agee for years. An exposition of the logic which justifies this research appears in the text of his paper, "The Sugar Planter Looks at Botany" published in 1932 (1).

In the summer of 1938 a definite field study of nitrogen in the cane plant was inaugurated by Agee at Waialua Agricultural Company, Ltd., and Ewa Plantation Company on the Island of Oahu. Cooperation in this study was contributed by several members of the plantation and Experiment Station staffs.

Efforts to locate distinctive zones in the cane plant, the analysis of which might yield information on its nutrient requirement, formed the early objective of the study. Various methods and procedures were developed as aids in this search, among which was a rapid chemical method for the analysis of leaf specimens to be obtained by ticket-punch procedure (10). Earlier studies by Q. H. Yuen and L. E. Davis, chemistry department of this Experiment Station, gave indications which suggested the feasibility of investigating the leaf-punch technic of sampling the growing plant. Further and more extensive field studies were undertaken. From results of these investigations, a standardized sampling procedure for an organized

foliar study of field canes was developed and the original rapid chemical method of analysis was modified to determine the total nitrogen content of the specimens on a moisture-free basis. The standardized procedure, as currently employed (April 1939) for leaf-punch collections, and the rapid chemical method of nitrogen determination will now be described.

Briefly stated, the samples are obtained with a ticket punch from localized areas of a definite group of leaves of a stalk of sugar cane. The punch cuts a disk 5 mm. in diameter. The leaf specimens are oven dried and analyzed for total nitrogen. The analytical procedure consists essentially of digesting the sample in a semi-micro Kjeldahl flask, using potassium sulfate and sulfuric acid containing a selenium catalyst. The sample, after digestion, is cooled and diluted with water. Strong alkali is added and the liberated ammonia is distilled into a water-jacketed absorption tube containing dilute sulfuric acid. After distillation and cooling, the solution containing the absorbed ammonia is made to a definite volume. Aliquots are taken for the colorimetric determination of nitrogen, using a specially prepared Nessler reagent with standard color solutions.

The procedure in detail follows:

1. Obtain samples in the field from growing cane.
2. Sample the leaves of the second, third and fourth "dewlap" (ligule) attachments on each stalk. The numbering starts from the top, counting the first *visible* dewlap nearest to the spindle as one. Punch two disks from each leaf at a point about midway from tip to base and about midway from margin to midrib, punching either or both blades.
3. Sample 10 stalks, obtaining 60 disks, or between 0.04 to 0.1 gram of dry tissue.
4. Transfer disks to a small tin box and cover securely.
5. Remove cover and dry in electric oven at 100°C. for 3 hours, or at 80°C. for 5 hours (or overnight). (Cover and receptacle may be nested.) When dry, remove cans from oven; replace covers. Place cans in a desiccator and cool for 15 minutes, or longer. Obtain dry weight of sample by transferring it to tared scoop and weighing on analytical balance. Record the dry weight.
6. Transfer the disks to a Kjeldahl flask of 300-ml. capacity.
7. A porous granule (about the size of half a corn grain) is dropped into the flask. About 0.3 gram (approximately  $\frac{1}{3}$  small teaspoonful) of special grade powdered potassium sulfate is added. Using a special pipette, 4 ml. of Reagent 15 Total N (sulfuric acid containing selenium) are transferred into the flask. The flask is rotated until every leaf disk is coated with the acid. The flask is then placed on a precision heater which has been preheated for at least 5 minutes and the digestion is thus continued for 30 minutes.

After this digestion period the flask is removed from the heater and cooled. One hundred ml. Reagent 18 Total N (distilled water) are then added, washing down the neck of the flask in the process. Fifteen ml. of Reagent 16 Total N (dilute sulfuric acid) are transferred to a calibrated test tube (200 x 29 mm.). The test tube is then immersed in a wide-mouth 500-ml. flask of water. The apparatus for distillation is set up. (For further description of the analytical procedure and reagents consult the paper (9) on rapid chemical method for the determination of total nitrogen in cane juice.) Add 20 ml. Reagent 17 Total N (strong alkali) to the Kjeldahl

flask and immediately connect with distillation assembly. Continue distillation until liquid in test tube reaches the calibrated mark and then remove from distillation apparatus.

8. The distillate, after cooling, is made up to 50-ml. volume with Reagent 18 Total N and mixed.

(a). In general, a mixture of 1 ml. distillate plus 5 ml. of Reagent 18 Total N and a 0.50-ml. distillate plus 5-ml. Reagent 18 Total N mixture will cover the range of nitrogen found in the sample. However, the table of readings to follow will cover any extremes, provided approximately 60-disk samples are taken.

9. (a). Using a special 1-ml. (calibrated to 0.1 ml.) Mohr pipette, transfer 1-ml. aliquots of the distillate to each of two comparison vials. Add 5 ml. Reagent 18 Total N with a special 5-ml. Mohr pipette. Add 1 ml. Reagent 6 N to each tube. Stopper and let stand 1 minute; mix if necessary. Then compare with color standards on the illuminator.

(b). Repeat procedure with another set of two vials, but use a 0.50-ml. aliquot of the distillate plus 5 ml. of Reagent 18 Total N.

(c). When proficiency in reading and matching color standards has been attained, the two different proportional mixtures may be made up at one time, developed, allowed to stand the 1-minute interval and then read.

(d). The other dilutions listed in the Table of Readings (0.25 ml. distillate plus 5 ml. Reagent 18 Total N and 1.5 ml. distillate plus 4.5 ml. Reagent 18 Total N) are only to be used when the 1-ml. and 0.5 ml. aliquots are unsatisfactory because of a too light or too dark color development. Standard tubes which give most satisfactory results are those between 3 and 6, inclusive.

10. Refer to the Table of Readings (Table I) for data on the calculation of the Percentage of Nitrogen in Leaf-Punch Samples. (The fractions between standard numbers of the table refer to the position of the unknown to its approximate matching between two adjacent standards.)

11. Refer to the Table of Factors for Dry Weight (Table II).

12. Factor times Reading = per cent Total N (dry basis). The average of the two readings multiplied by the Factor for Dry Weight will give the result for the analysis of the sample.

TABLE I

TABLE OF READINGS FOR NITROGEN DETERMINATION IN LEAF-PUNCH  
SAMPLES WHERE ENTIRE SAMPLE IS DISTILLED

(Dilutions below refer to Treatment of Distillate)

Standard No.	Dilution			
	1 ml. + 5	0.50 ml. + 5	0.25 ml. + 5	1.5 ml. + 4.5
1	0.012	0.022	0.042	0.008
.25	0.018	0.033	0.063	0.012
.50	0.024	0.044	0.084	0.016
.75	0.030	0.055	0.105	0.020
2	0.036	0.066	0.126	0.024
.25	0.042	0.077	0.147	0.028
.50	0.048	0.088	0.168	0.032
.75	0.054	0.099	0.189	0.036
3	0.060	0.110	0.210	0.040
.25	0.066	0.121	0.231	0.044
.50	0.072	0.132	0.252	0.048
.75	0.078	0.143	0.273	0.052
4	0.084	0.154	0.294	0.056
.25	0.090	0.165	0.315	0.060
.50	0.096	0.176	0.336	0.064
.75	0.102	0.187	0.357	0.068
5	0.108	0.198	0.378	0.072
.25	0.117	0.215	0.410	0.078
.50	0.126	0.231	0.442	0.084
.75	0.135	0.248	0.473	0.090
6	0.144	0.264	0.504	0.096
.25	0.153	0.281	0.536	0.102
.50	0.162	0.297	0.567	0.108
.75	0.171	0.314	0.598	0.114
7	0.180	0.330	0.630	0.120
		0.358	0.682	0.130
				0.140

### ERRATUM

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First line on page 128 should be first line on page 127.

TABLE II  
TABLE OF FACTORS FOR DRY WEIGHT

The following factors are obtained by the formula:

$$\text{Factor (F)} = \frac{1}{\text{Dry weight of sample}}$$

Dry weight	Factor	Dry weight	Factor	Dry weight	Factor
0.040	25.0	0.060	16.7	0.080	12.5
0.041	24.4	0.061	16.4	0.081	12.3
0.042	23.8	0.062	16.1	0.082	12.2
0.043	23.2	0.063	15.9	0.083	12.0
0.044	22.7	0.064	15.6	0.084	11.9
0.045	22.2	0.065	15.4	0.085	11.8
0.046	21.7	0.066	15.1	0.086	11.6
0.047	21.2	0.067	14.9	0.087	11.5
0.048	20.8	0.068	14.7	0.088	11.4
0.049	20.4	0.069	14.5	0.089	11.2
0.050	20.0	0.070	14.3	0.090	11.1
0.051	19.6	0.071	14.1	0.091	11.0
0.052	19.2	0.072	13.9	0.092	10.9
0.053	18.9	0.073	13.7	0.093	10.8
0.054	18.5	0.074	13.5	0.094	10.6
0.055	18.2	0.075	13.3	0.095	10.5
0.056	17.8	0.076	13.2	0.096	10.4
0.057	17.5	0.077	13.0	0.097	10.3
0.058	17.2	0.078	12.8	0.098	10.2
0.059	16.9	0.079	12.6	0.099	10.1
0.060	16.7	0.080	12.5	0.100	10.0

*Example:*

- (a) Dry weight of sample = 0.0812 gram
- (b) Distillate analysis, Reading:  
Dilution 1 and 5 = 0.180 Reading  
Dilution  $\frac{1}{2}$  and 5 = 0.176 Reading
- (c) Referring to Table of Factors for Dry Weight,  
0.081 gram, Factor = 12.3
- (d) Per cent total N = Factor times Reading  
1 ml.+5 ml. dilution— $12.3 \times 0.180 = 2.21\%$  T.N.  
0.5 ml.+5 ml. dilution— $12.3 \times 0.176 = 2.16\%$  T.N.  $\left. \vphantom{\begin{array}{l} 1 \text{ ml.+5 ml. dilution} \\ 0.5 \text{ ml.+5 ml. dilution} \end{array}} \right\} 2.19\%$
- (e) or averaging the two readings:  
 $\left. \begin{array}{l} 0.180 \\ 0.176 \end{array} \right\} 0.178 \text{ (average reading)}$   
and multiplying by Factor:  
 $0.178 \times 12.3 = 2.19\%$  (result for the sample).

#### *Discussion on the Field Sampling Procedure:*

The procedure is based on sampling a definite grouping of leaves of the plant, but with random selection of stalks. In general, the object consists in obtaining a representative sample of cane growing in a field. In a stand of cane there may exist different orders of stalks, including tasseled and untasseled canes. Experience thus far has indicated that for consistency of results only the untasseled cane should be sampled. In selecting stalks for sampling, as much of the primary growth is included as may be consistent with the principle of random selection.

To insure reliability of results two composite samples are obtained from each field. Two sampling stations are thus established per field, each station comprising an area of about 25 feet square ( $25' \times 25'$ ). Sixty leaf-punch disks constitute a sample. Two disks are taken from each leaf in a region located about halfway along the blade and about one-half the distance between the midrib and the outer edge of the leaf. The two disks may be spaced about an inch apart along the blade. The critical sampling zone comprises that portion of the leaf system representing the leaves attached to the second, third and fourth visible dewlaps (ligules), counting from the top of the plant. These three designated leaves appear to comprise that portion of the plant which will yield reliable information concerning the "nitrogen index." (The nitrogen content percentage of the specimen has been termed the "nitrogen index" by Mr. Agee.)

Sampling may start with cane about three months of age. For young cane, not yet head high (less than 6 feet from base to top of foliage) only the second leaf is punched. As the plant grows, the third and fourth dewlap leaves are included. The full complement of second, third and fourth dewlap leaves may be sampled generally after the cane has reached or exceeded the above designated height. When the full complement of leaves comprises the material to be sampled, ten stalks are selected for the purpose in each station. For young cane, a sufficient number of stalks are selected to insure a 60-disk sample.

Where it is desired to obtain leaf specimens before the plant reaches an age of three months, cane growing not longer than a period of two months may be sampled. In such a case only the first dewlap leaf should be punched and this obvious deviation from the regular sampling order should be recorded with the analytical data accruing from the analysis.

#### *Accuracy and Precision of Analytical Method:*

*Accuracy:* In the regular laboratory method for analyzing plant material, the sample used in analysis ranges from one to five grams dry matter. The macro-Kjeldahl procedure is used and the resulting ammoniacal distillate is titrated with standard acid. In the rapid chemical method only approximately 0.060 to 0.080 gram of dry matter is used for each determination. The resulting distillate is analyzed with a colorimetric procedure. The final concentration of the distillate used in colorimetric analysis ranges from 0.4 to 8 parts-per-million nitrogen so that the dilution factor is exceedingly large. In spite of the differences in quantities of sample and methods of analysis, the accuracy of the rapid method is extremely close in comparison with the macro method. For leaf-punch analysis the usual range of nitrogen content is between one and three per cent on the dry-weight basis. The accuracy for this type of analysis is found to be within at least 90 per cent of the regular laboratory method. It has been found that for cane juice analysis and certain biological solutions where the composition is between 0.010 and 0.200 per cent of total nitrogen the results obtained by the two methods are practically identical.

*Precision:* In an organized nitrogen study of the sugar cane leaf system, the data obtained are examined from the standpoint of time or age relationship. The precision of the method and the agreement between results of duplicate analysis become increasingly important. The significance of the difference between nitrogen

indices obtained during the growth period of the crop is dependent upon the reliability of the method and the accuracy of collecting samples and analyzing them.

Surprisingly close agreement has been noted thus far in analysis of duplicate samples from many fields. These duplicates always originate in widely separated stations. Many varieties and different ages of cane have been examined for the effect on inherent nitrogen variability. While H 109 gave the closest agreement, other varieties like 31-1389, POJ 2878, H 8965 and 27-8101 are not extremely variable in this respect. In H 109 cane growing uniformly in a field, it appears that the difference between duplicate determinations of nitrogen index values is usually found at minimum or not exceeding 0.20 per cent total nitrogen. For other varieties similarly checked, the difference may exceed 0.20 per cent but the variability usually falls within 0.40 per cent total nitrogen. Thus for the variety H 109 it may appear that where the difference between the averages of two duplicate sets of analyses determined at different intervals of growth does not exceed 0.20 per cent total nitrogen, the variation may be due to analytical error. However, where the difference exceeds 0.40 per cent, it may be accepted as reliably significant.

The variation in nitrogen index values of H 109 was studied by one of us in a plant crop. Eight plots of 3 lines each, at Makiki, were located in widely separated parts of a quarter-acre field divided into sixty plots. Leaf-punch samples were taken from each plot and analyzed separately. The data resulting from analyses made on leaf-punch specimens from the eight plots were averaged and the probable error of the mean (PEM) and coefficient of variation per sampling were determined. The first samples were taken at the age of six weeks and at each monthly and sometimes bi-weekly intervals. Eleven sets of samples were secured during a crop interval of 8 months. The average percentage content of each set ranged from 1.72 per cent to 2.43 per cent. The probable error of the mean for each set per sampling period fluctuated between 0.02 and 0.05 per cent total nitrogen. The coefficients of variation ranged from 2.3 per cent to 6.5 per cent, with the majority within 4 per cent. Similar results were obtained with POJ 2878 in the same field.

*A preliminary presentation is offered of the results of the leaf-punch nitrogen study so far concluded in the effort to establish a physiological basis for the analysis in controlling nitrogen fertilization of sugar cane. The investigation embraces (a) a study of the relationship between nitrogen content of the leaf disks and the growth of sugar cane in pots, (b) a study of field results, and (c) a presentation of a tentative procedure which may aid in the practical control of nitrogen fertilization.*

#### POT EXPERIMENT ON CANE GROWTH

It is generally accepted, we believe, that in many plants and especially sugar cane (as shown in many other experiments) the nitrogen content of the foliage fluctuates from a high level during the young stages of growth to a low value at maturity. In order that any method of leaf analysis may be applicable to the purpose of fertilization control, a relationship should exist between the nitrogen content determined at any time of growth and the stage of the plant development under study at that time. A pot experiment was conducted to ascertain if such a positive relationship, if any, existed and, if so, to what extent and to what degree the course of nitrogen variation fluctuated in the leaf system concurrently with growth.

*Experimental Plan:*

In this experiment pregerminated H 109 cane shoots were grown in Mitscherlich pots, using a Manoa acid soil from Field 5. Sufficient phosphate and potash (9.0 grams  $P_2O_5$  from solution of superphosphate, and 3.0 grams  $K_2O$  from potash sulfate) were mixed with  $4\frac{1}{2}$  kilograms of air-dry soil at the time of potting to eliminate the deficiency of these nutrients in the experiment. Nitrogen, therefore, became the limiting factor under study. Nitrogen treatments provided for three comparative levels of fertility (deficiency, sufficiency, and excess) with additional check pots to which no nitrogen fertilizers had been added.

The experiment consisted of 24 pots. Two pots did not receive nitrogen. The remaining pots were divided into two series of 11 each. One series received nitrogen as sulfate of ammonia and the other as nitrate of soda.

The treatments furnished the following amounts of nitrogen: 0, 1,  $1\frac{1}{2}$ , 2, and 3 grams of nitrogen to each pot of two stalks. Within each series the nitrogen was split into two applications, excepting the  $1\frac{1}{2}$ -gram treatment. For the split applications, one-half of each treatment was applied two weeks after potting. Variable times of application entered into the addition of the remaining half. For one sub-series, the remaining half was applied as soon as R.C.M. (rapid chemical methods) analysis showed the first application to be depleted from the soil. For another sub-series, the remaining half was applied when the plants showed definite yellowing of foliage.

Summarizing: the experiment was divided into two series: (a) sulfate of ammonia treatment, and (b) nitrate of soda. For each series, nitrogen applications were made of 0, 1,  $1\frac{1}{2}$ , 2, and 3 grams of nitrogen in the following manner:

2 pots no nitrogen treatment—same pots are common for each series

2 pots 1-gram nitrogen treatment (deficiency level)

$\frac{1}{2}$  gram at 2 weeks

$\frac{1}{2}$  gram when R.C.M. soil analysis showed depletion of first application

2 pots 1-gram nitrogen treatment

$\frac{1}{2}$  gram at 2 weeks

$\frac{1}{2}$  gram upon yellowing of foliage

2 pots 2-gram nitrogen treatment (sufficiency level)

1 gram at 2 weeks

1 gram when R.C.M. soil analysis showed depletion of first application

2 pots 2-gram nitrogen treatment

1 gram at 2 weeks

1 gram upon yellowing of foliage

2 pots 3-gram nitrogen treatment (excess level)

$1\frac{1}{2}$  grams at 2 weeks

$1\frac{1}{2}$  grams when R.C.M. soil analyses showed depletion of first application

1 pot  $1\frac{1}{2}$ -gram nitrogen treatment, one application at 2 weeks.

Two plants only were allowed to grow in each pot. As soon as suckers emerged from the soil they were removed. Periodic sampling of the leaves of the two plants per pot were made at frequent intervals by the leaf-punch technic. At the younger stages only the first fully opened leaf was sampled. As the growth of the plants progressed with the development of a complete leaf system, sampling was conducted



in a manner comparable to the regular procedure previously described and confined to the prescribed grouping of leaves.

The disk specimens were analyzed by the regular rapid chemical method developed for this work. Due to the smaller quantity of material taken for analysis, the method was modified only to the extent of facilitating colorimetric reading and calculation of data. Results expressed as percentage of total nitrogen were reported on both the green- and dry-weight bases of the samples.

Commencing with six weeks after potting and at periodic intervals coincident with leaf punching, linear growth measurements were made of each stalk of cane, measuring from a fixed base level on the pot to the uppermost visible dewlap of the stalk.

Soil analyses were made only up to the depletion of the initial application for each treatment in order to control the addition of the second half as planned.

### *Results:*

The data presented will cover only the nitrogen percentages of the leaf samples and the growth measurements. They represent averages for each treatment. Soil data obtained only for control will not be reported. Tables III, IV, VI, and VII give the nitrogen percentages of the leaf-punch samples on both the green- and dry-weight bases. The data on accumulative elongation of the stalk for each treatment are presented in Tables V and VIII.

Due to limited greenhouse facilities during the conduct of the experiment, the series of nitrate of soda pots was often subjected to shading which did not occur to any great extent in the sulfate of ammonia series. Shading favored greater elongation in the nitrate pots. While the data of the two series are not exactly alike, they are comparable and lead to similar and parallel conclusions. For purposes of simplification and clarity, the results of the sulfate of ammonia series only will be discussed later.

### *Reporting of Data:*

Differences of opinion exist as to the manner of expressing the results of analysis of plant materials for inorganic constituents. There are those who favor reporting results on the fresh- or green-weight basis of the samples as being best indicative of metabolic processes. Others favor a dry-weight basis as being more nearly correct due to the elimination of dilution of a small quantity of nutrients by a large proportion of moisture which is associated with most plant materials. The general conclusion at present appears to be that the procedure should be followed which is best suited for individual purposes. From results of this investigation it appears that expression of nitrogen data on the dry-weight basis is best suited for our purpose.

TABLE III  
PER CENT TOTAL NITROGEN IN LEAF DISKS (DRY BASIS) AT  
PROGRESSIVE INTERVALS

Sulfate of Ammonia Treatments

Applications:  $\frac{1}{2}$  of total quantity applied at 2 weeks

Remaining half applied (underlined):

(a) When R.C.M. soil analysis showed depletion

(b) Upon yellowing of foliage

Weeks	0 Check	<u>1 gm. N</u>		<u>2 gms. N</u>		3 gms. N	1½ gms. N
		(a)	(b)	(a)	(b)	(a)	at 2 wks.
Start	1.97						
2.....	3.42	2.81	2.81	2.72	2.72	2.60	2.60
4.....	3.39	3.30	3.30	3.16	3.16	3.64	3.64
6.....	2.84	3.44	3.44	3.44	3.44	3.40	3.40
8.....	1.46	2.64	2.64	2.52	2.52	2.56	2.56
10.....	1.43	<u>2.46</u>	2.46	2.49	2.49	2.97	2.97
12.....	1.32	<u>2.43</u>	1.69	2.34	2.34	2.54	2.54
14.....	1.09	1.80	1.62	<u>2.14</u>	2.14	2.48	2.48
16.....	1.09	1.52	<u>1.27</u>	<u>2.36</u>	1.67	2.05	2.05
18.....	0.74	1.17	<u>1.39</u>	1.66	1.14	<u>1.50</u>	1.50
20.....	0.86	1.17	1.52	1.74	<u>1.22</u>	<u>2.08</u>	1.57
22.....	0.90	1.04	1.46	1.38	<u>1.66</u>	1.90	1.25
24.....	0.78	1.08	1.08	1.14	1.44	1.66	0.95
26.....	1.06	1.02	1.04	1.16	1.26	1.47	1.06
28.....	0.93	0.80	0.84	0.92	1.12	1.31	0.94
30.....	0.90	0.75	0.81	0.96	0.94	1.10	0.84
32.....	0.97	0.73	0.72	0.74	0.99	0.98	0.80
36.....	0.85	0.67	0.64	0.74	0.78	0.87	0.69

TABLE IV  
PER CENT TOTAL NITROGEN IN LEAF DISKS (GREEN BASIS) AT  
PROGRESSIVE INTERVALS

Sulfate of Ammonia Treatments

Applications:  $\frac{1}{2}$  of total quantity applied at 2 weeks

Remaining half applied (underlined):

(a) When R.C.M. soil analysis showed depletion

(b) Upon yellowing of foliage

Weeks	0. Check	1 gm. N		2 gms. N		3 gms. N	1½ gms. N
		(a)	(b)	(a)	(b)	(a)	at 2 wks.
Start	0.47						
2.....	1.20	1.03	1.03	0.97	0.97	0.83	0.83
4.....	1.39	1.24	1.24	1.23	1.23	1.50	1.50
6.....	0.80	1.01	1.01	1.16	1.16	1.07	1.07
8.....	0.53	0.99	0.99	0.92	0.92	0.93	0.93
10.....	0.47	<u>0.79</u>	0.79	0.80	0.80	0.92	0.92
12.....	0.47	0.76	0.50	0.70	0.70	0.79	0.79
14.....	0.40	0.56	0.52	<u>0.66</u>	0.66	0.79	0.79
16.....	0.40	0.52	<u>0.45</u>	<u>0.80</u>	0.55	0.68	0.68
18.....	0.30	0.42	<u>0.50</u>	0.62	<u>0.36</u>	<u>0.50</u>	0.50
20.....	0.28	0.37	0.48	0.55	<u>0.38</u>	0.66	0.55
22.....	0.31	0.34	0.48	0.44	<u>0.57</u>	0.64	0.42
24.....	0.30	0.38	0.36	0.40	0.53	0.54	0.32
26.....	0.37	0.35	0.34	0.37	0.40	0.46	0.36
28.....	0.34	0.29	0.29	0.32	0.35	0.46	0.34
30.....	0.32	0.26	0.29	0.33	0.31	0.38	0.31
32.....	0.35	0.27	0.28	0.28	0.35	0.36	0.30
36.....	0.31	0.25	0.25	0.28	0.29	0.31	0.26

TABLE V  
ACCUMULATIVE STEM ELONGATION (IN CENTIMETERS) PER POT OF  
TWO STALKS AT PROGRESSIVE INTERVALS

Sulfate of Ammonia Treatments

Nitrogen applications:  $\frac{1}{2}$  of total quantity applied at 2 weeks  
Remaining half applied (underlined):

(a) When R.C.M. soil analysis showed depletion

(b) Upon yellowing of foliage

Weeks	0 Check	1 gm. N		2 gms. N		3 gms. N	1½ gms. N
		(a)	(b)	(a)	(b)	(a)	at 2 wks.
6.....	0.9	2.7	2.7	3.7	3.7	2.4	2.4
8.....	1.9	12.8	12.8	10.6	10.6	7.7	7.7
10.....	2.3	<u>19.6</u>	19.6	18.4	18.4	14.7	14.7
12.....	2.6	<u>28.0</u>	23.8	28.8	28.8	25.9	25.9
14.....	2.6	35.6	26.1	<u>35.7</u>	35.7	34.4	34.4
16.....	2.6	39.8	<u>27.5</u>	<u>48.4</u>	41.3	43.7	43.7
18.....	2.6	<u>42.1</u>	40.0	59.0	<u>46.4</u>	<u>51.1</u>	51.1
20.....	2.7	43.9	48.2	64.8	<u>48.4</u>	<u>57.1</u>	57.2
22.....	2.7	44.5	51.0	68.5	<u>54.6</u>	68.1	62.2
24.....	2.7	46.1	52.4	71.1	66.3	71.9	64.2
26.....	2.8	46.7	54.0	76.5	74.6	77.9	64.3
28.....	3.8	47.0	54.6	82.1	81.0	87.1	64.9
30.....	3.9	47.4	55.6	85.5	83.3	92.1	65.0
32.....	4.2	48.1	56.3	87.3	86.4	98.7	67.7
34.....	4.7	48.3	56.5	88.9	90.2	103.6	68.9
36.....	5.9	48.9	59.5	95.5	98.8	112.8	70.0
38.....	7.3	50.1	61.8	101.1	107.4	121.5	75.3
40.....	7.9	50.7	63.2	104.9	110.7	124.6	76.1
42.....	9.0	50.9	64.4	110.8	115.0	128.2	76.2
44.....	9.8	51.0	65.8	113.5	118.8	129.6	76.2
46.....	11.2	51.4	68.7	115.5	121.5	133.1	77.7
48.....	11.5	52.4	70.4	117.0	125.3	137.3	78.2
50.....	12.1	53.0	71.4	117.4	127.5	138.9	79.8
52.....	12.9	53.2	71.7	117.7	129.2	140.3	80.5

TABLE VI  
PER CENT TOTAL NITROGEN IN LEAF DISKS (DRY BASIS) AT  
PROGRESSIVE INTERVALS

Nitrate of Soda Treatments

Applications:  $\frac{1}{2}$  of total quantity applied at 2 weeks

Remaining half applied (underlined):

(a) When R.C.M. soil analysis showed depletion

(b) Upon yellowing of foliage

Weeks	.0 Check	1 gm. N		2 gms. N		3 gms. N	1½ gms. N
		(a)	(b)	(a)	(b)	(a)	at 2 wks.
Start	1.97						
2.....	3.42	2.66	2.66	2.63	2.63	2.51	2.51
4.....	3.39	2.82	2.82	3.33	3.33	3.29	3.29
6.....	2.84	3.34	3.34	3.37	3.37	3.20	3.20
8.....	1.46	2.56	2.56	2.47	2.47	2.54	2.54
10.....	1.43	<u>2.68</u>	2.68	2.86	2.86	3.48	3.48
12.....	1.32	<u>2.30</u>	2.02	2.35	2.35	2.82	2.82
14.....	1.09	2.49	2.11	<u>2.36</u>	2.36	2.67	2.67
16.....	1.09	1.96	<u>1.48</u>	<u>2.31</u>	2.08	2.35	2.35
18.....	0.74	1.47	<u>1.66</u>	1.92	1.50	1.90	1.90
20.....	0.86	1.38	1.69	1.84	<u>1.48</u>	2.02	1.33
22.....	0.90	1.06	1.36	1.60	<u>1.76</u>	1.86	1.10
24.....	0.78	0.88	1.03	1.46	1.45	1.68	1.00
26.....	1.06	0.81	0.98	1.27	1.37	1.62	0.98
28.....	0.93	0.84	0.86	0.99	1.08	1.10	0.82
30.....	0.90	0.80	0.84	1.05	1.21	1.24	1.04
32.....	0.97	0.73	0.76	0.98	0.85	0.97	0.91
36.....	0.85	0.66	0.68	0.75	0.73	0.83	0.75

**TABLE VII**  
**PER CENT TOTAL NITROGEN IN LEAF DISKS (GREEN BASIS) AT**  
**PROGRESSIVE INTERVALS**

Nitrate of Soda Treatments

Applications:  $\frac{1}{2}$  of total quantity applied at 2 weeks  
 Remaining half applied (underlined):

(a) When R.C.M. soil analysis showed depletion  
 (b) Upon yellowing of foliage

Weeks	0 Check	$\underbrace{1 \text{ gm. N}}_{\substack{(a) \quad (b)}}$		$\underbrace{2 \text{ gms. N}}_{\substack{(a) \quad (b)}}$		3 gms. N (a)	1½ gms. N at 2 wks.
Start	0.47						
2.....	1.20	0.97	0.97	0.94	0.94	0.79	0.79
4.....	1.39	1.16	1.16	1.38	1.38	1.38	1.38
6.....	0.80	0.99	0.99	0.95	0.95	0.93	0.93
8.....	0.53	0.90	0.90	0.87	0.87	0.89	0.89
10.....	0.47	<u>0.82</u>	0.82	0.85	0.85	1.01	1.01
12.....	0.47	<u>0.73</u>	0.62	0.70	0.70	0.85	0.85
14.....	0.40	0.75	0.66	<u>0.71</u>	0.71	0.81	0.81
16.....	0.40	0.68	<u>0.49</u>	<u>0.75</u>	0.65	0.76	0.76
18.....	0.30	0.52	<u>0.56</u>	0.62	0.47	<u>0.61</u>	0.61
20.....	0.28	0.42	0.53	0.55	<u>0.42</u>	<u>0.60</u>	0.39
22.....	0.31	0.34	0.44	0.52	<u>0.56</u>	0.60	0.36
24.....	0.30	0.30	0.34	0.48	0.48	0.58	0.35
26.....	0.37	0.28	0.33	0.42	0.46	0.52	0.32
28.....	0.34	0.33	0.28	0.39	0.42	0.41	0.32
30.....	0.32	0.28	0.28	0.37	0.40	0.42	0.38
32.....	0.35	0.27	0.26	0.32	0.30	0.34	0.34
36.....	0.31	0.23	0.26	0.28	0.27	0.32	0.29

TABLE VIII  
ACCUMULATIVE STEM ELONGATION (IN CENTIMETERS) PER POT OF  
TWO STALKS AT PROGRESSIVE INTERVALS

Nitrate of Soda Treatments

Nitrogen applications:  $\frac{1}{2}$  of total quantity applied at 2 weeks  
Remaining half applied (underlined):

(a) When R.C.M. soil analysis showed depletion

(b) Upon yellowing of foliage

Weeks	0 Check	1 gm. N		2 gms. N		3 gms. N	$1\frac{1}{2}$ gms. N at 2 wks.
		(a)	(b)	(a)	(b)	(a)	
6.....	0.9	3.8	3.8	3.4	3.4	2.9	2.9
8.....	1.9	11.7	11.7	10.7	10.7	7.8	7.8
10.....	2.3	17.6	17.6	17.6	17.6	13.2	13.2
12.....	2.6	23.9	24.6	28.7	28.7	27.1	27.1
14.....	2.6	30.1	26.4	36.2	36.2	34.6	34.6
16.....	2.6	38.9	27.4	47.0	45.3	47.5	47.5
18.....	2.6	45.7	43.6	61.0	53.1	60.2	60.2
20.....	2.7	50.7	52.9	73.0	58.9	65.3	63.0
22.....	2.7	52.5	56.6	82.2	65.7	69.0	66.7
24.....	2.7	53.1	57.6	90.6	79.8	78.0	67.6
26.....	2.8	53.8	59.2	98.6	91.0	88.5	72.3
28.....	3.8	56.0	61.1	104.3	101.7	98.9	76.9
30.....	3.9	56.8	62.3	107.9	110.2	106.2	80.7
32.....	4.2	57.2	64.1	109.8	120.6	112.4	84.4
34.....	4.7	57.3	65.5	110.5	126.8	116.3	86.7
36.....	5.9	59.7	72.4	113.5	139.9	124.7	92.1
38.....	7.3	61.6	75.9	116.5	152.7	133.1	98.9
40.....	7.9	62.2	77.3	117.5	158.1	137.7	104.2
42.....	9.0	62.2	77.9	117.5	161.7	139.9	107.1
44.....	9.8	62.2	80.7	117.5	162.3	141.6	107.7
46.....	11.2	63.3	83.5	118.8	163.3	146.7	110.1
48.....	11.5	65.4	87.6	120.0	165.7	151.8	111.4
50.....	12.1	66.2	90.0	120.2	168.0	154.0	111.9
52.....	12.9	67.4	91.5	120.6	168.5	155.7	112.3

An inspection of Tables III and IV will reveal that similar trends may be noted for nitrogen values determined at progressive intervals and expressed on both bases. However, values on the dry-weight basis are much higher than those calculated on green weights. It appears, therefore, that greater differences may be detected with the dry-weight data. A tabulation of the 2-gram nitrogen from sulfate of ammonia treatment is given to illustrate this point.

#### PER CENT TOTAL NITROGEN IN LEAF SAMPLES

Green-weight basis	Dry-weight basis
.97	2.72
.92	2.52
.80	2.49
.80	2.36
.70	2.34
.66	2.14
.62	1.66
.55	1.74
.44	1.38
.40	1.14
.37	1.16
.33	0.96
.32	0.92
.28	0.74

From the above data it is apparent that the differences between the highest and the lowest values are 0.69 per cent total nitrogen for the green-weight basis and 1.98 per cent for the dry basis. A wider range of fluctuation is possible for the dry-weight values. Its effect on showing differences is apparent on comparing a few data. For instance, if 0.66 per cent and 0.62 per cent green-weight basis are compared, the difference between the two values is 0.04 per cent, which may be too small to be significant for practical and routine operation. However, with their corresponding dry-weight values, the difference between 2.14 per cent and 1.66 per cent becomes 0.48 per cent total nitrogen and may be accepted as reliable. For a narrow fluctuation between 0.40 per cent and 0.55 per cent on the green-weight basis, their dry-weight values become 1.14 per cent and 1.74 per cent.

Any changes in the moisture content of the leaf samples as influenced by the time of day sampled, age of cane, recent rainfall or irrigation and seasonal differences may have an effect on the variation of the nitrogen content on the green-weight basis of the samples collected. It is thus difficult to determine whether fluctuations are due to normal changes of growth or whether they are due primarily to differences in moisture content of the samples. However, when the results are expressed on the dry-weight basis, at least one source of fluctuation is eliminated. The effect of rainfall on the nitrogen content has been definitely shown to change the fresh-weight results without affecting the dry-weight values. In this regard a study of the effect of rainfall by Fagan has been reported by Nishimura and Hance (13).

In addition to the absorbed moisture, the extraneous moisture adhering to the leaf from dew and showers make sampling difficult when results are to be expressed on the green-weight basis. Thus, in addition to physiological differences noted, the



difficulties encountered in an actual field procedure makes reporting on a green-weight basis undesirable and less practical. For these reasons the analytical data determined on the organized leaf-punch study, using a standardized sampling and analytical procedure, are reported on the moisture-free basis. In subsequent discussions, the data under consideration will refer to cane leaf nitrogen index values on the moisture-free basis. In Figs. 1, 2 and 3, nitrogen curves on the green-weight basis are included for inspection only.

### *Discussion of Results:*

*The Limits of Nitrogen Percentage Values Determined by Leaf Punch During Cane Growth:* Graphs of nitrogen indices are obtained by plotting the percentages of total leaf nitrogen on the ordinate (Y-axis) and the time or age interval on the abscissa (X-axis). In Figs. 1, 2 and 3 are presented the growth and nitrogen data for the check pots and those for the sulfate of ammonia treatment. The graphs of the dry-weight nitrogen data are typical of those obtained from the field for cane leaf analysis carried out by this method of study.

Examining the graph for the check pots it may be noted that from an initial figure of 1.97 per cent total nitrogen determined at the time of potting, the value increased to 3.42 per cent two weeks after potting. This value represents the highest obtained for these check pots. The curve dropped progressively with growth from this highest point to a low of about 1 per cent.

In the fertilized pots the nitrogen value two weeks after potting rose from 1.97 per cent to about 2.75 per cent, then to a high of 3.4 per cent two weeks after fertilization. From then on the curve dropped progressively with growth to a low of approximately 1 per cent and fluctuated between this value and 0.8 per cent.

In general, it appears that the highest values are obtained during the very young stage of growth of the cane plant. The curve then drops progressively. However, the slope of the curve depends to a certain extent upon the *time of application* and the quantity of nitrogen applied. From results of this experiment and subsequent pot and field studies, the maximum reached for percentage total nitrogen, irrespective of variety, is about 3 per cent. However, this value is usually reached prior to the age of three months and depends to a large extent upon the fertility of the soil with respect to all plant foods and the amount of nitrogen fertilization added. For field crops the highest value is closer to 2.6 per cent. The lowest value to which the nitrogen index is reduced in a normal crop may be placed at 1 per cent. Thus, the maximum range of fluctuation to be found in leaf-punch analysis is between 3 per cent and 1 per cent, regardless of variety or fertilization. That is, irrespective of whether the crop received no nitrogen or whether it was fertilized with 400 pounds of this nutrient, the leaf nitrogen values will fluctuate within these limits. The time required for the value to drop from the maximum to the minimum is, to a large degree, influenced by the quantity of nitrogen applied and absorbed by the plant. This is illustrated in Fig. 4.

In Fig. 4 nitrogen percentage curves are presented as representing the check and fertilized pots. These graphs show clearly that the time required for each curve to reach a minimum is dependent upon the amount of nitrogen received by the plants.

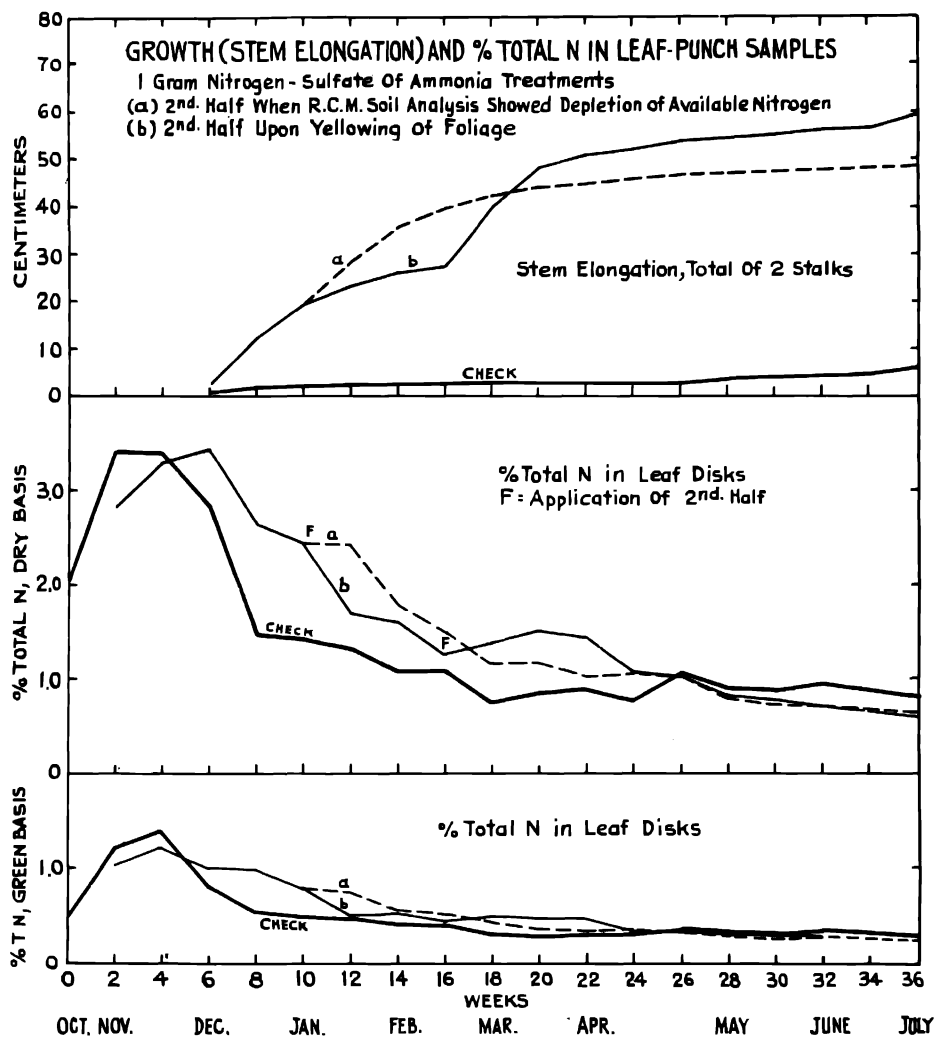


Fig. 1

For the check or no-nitrogen pots, the curve dropped to the minimum level within 14 weeks after potting. For the others it ranged from 20 to 32 weeks, depending upon the fertilization.

*Effect of Fertilization Upon Trend of Curve:* While the nitrogen indices may fluctuate within the limits stated, it is desirable to examine the data to determine to what extent these values may reflect differential applications of nitrogen. From a study of the fertilized pots in Fig. 4 it appeared that during the start of the crop the nitrogen indices were alike for all treatments. However, as growth progressed different values were obtained for the treatments at a given instant. This is to be expected if the interval required for the lowering to a minimum value is dependent upon the amount of nitrogen applied and growth of the crop. (It will be noted that the fluctuations occurred within or nearly within the previously stated range of values.)

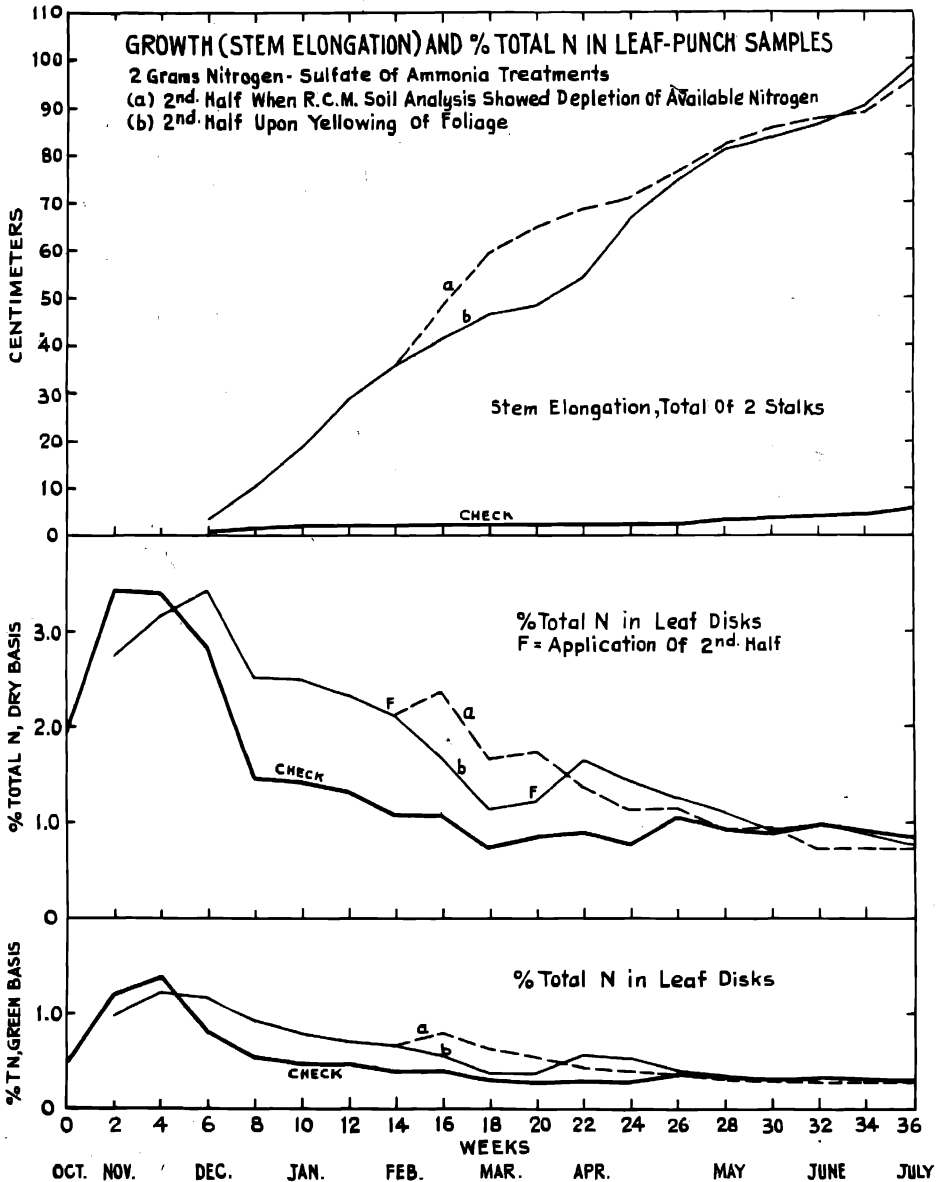


Fig. 2

The above discussion is based on data obtained in a pot experiment where only the original or primary stalk was developed. However, under field conditions where all orders of stalks are allowed to develop in one stool of cane, such differences in leaf nitrogen content may not become as readily apparent. Under field conditions, as the amount of nitrogen is increased for a given area, increase in tillering is usually obtained in addition to a development of larger leaf area per stalk. These increases in plant development may operate to equalize the excess which can accumulate if the primary stalk only is allowed to grow.

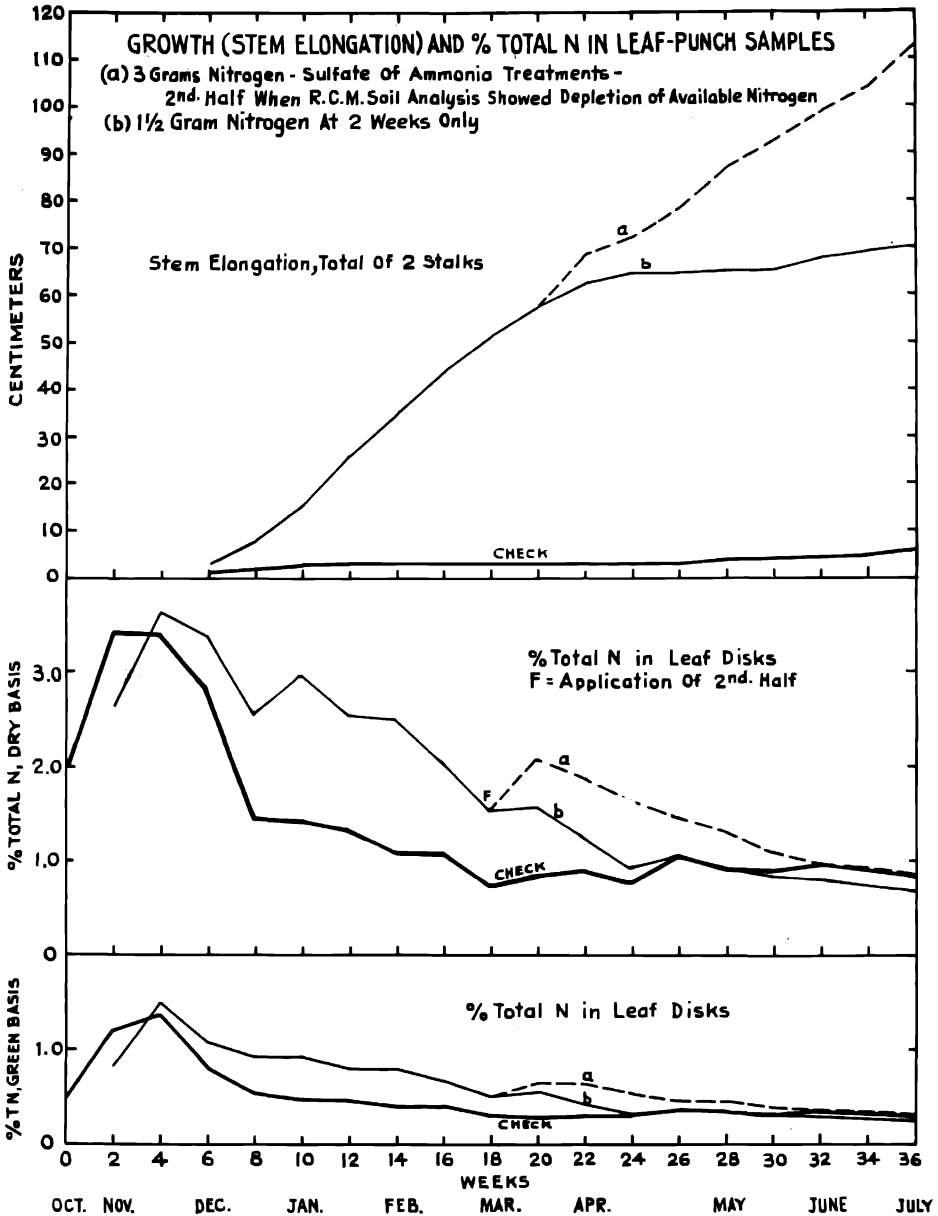


Fig. 3

In general field practice, the nitrogen requirement for a crop is usually split into several applications and applied at different times. It is desirable to know in what manner nitrogen applications may affect the index level shortly following fertilization.

The pot experiment under study provided for the conditions of a "time-of-application" and an "amounts-of-nitrogen" test. The solution of this question, therefore, may become apparent from an examination of Figs. 1, 2, 3 and 4. The results appear to indicate that fertilization may either (a) raise the nitrogen level

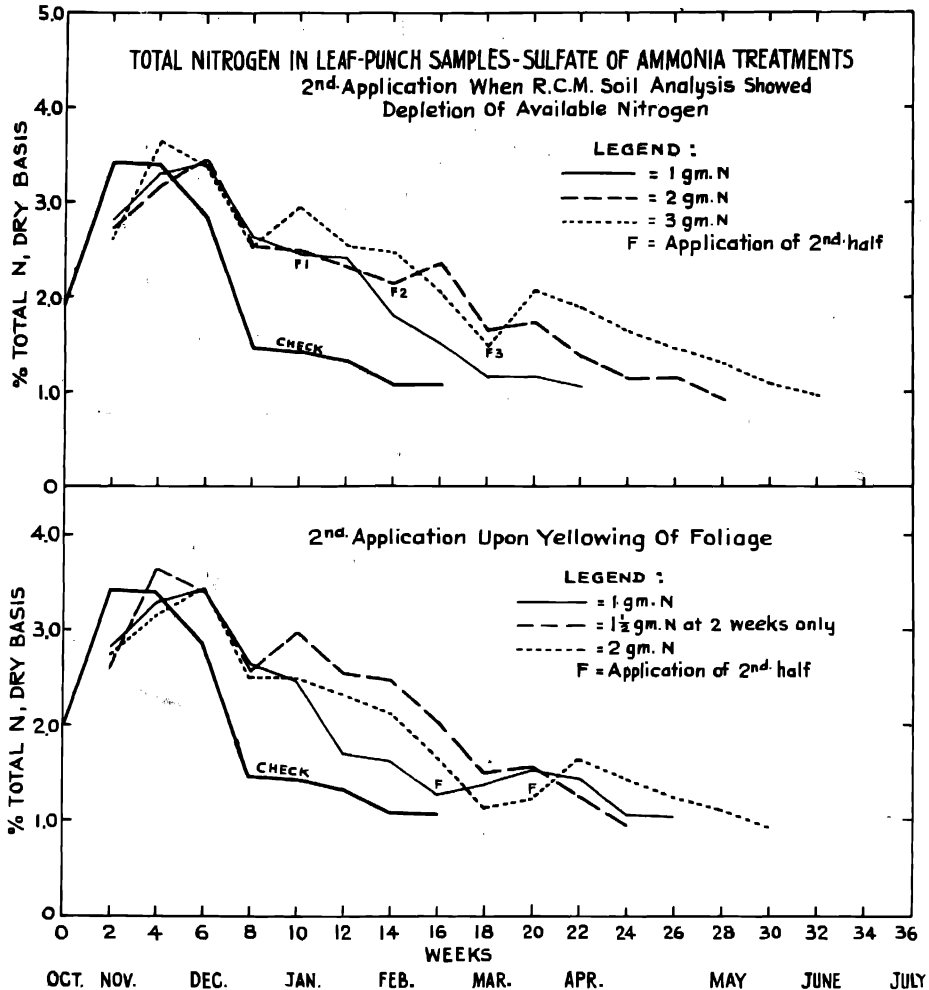


Fig. 4

above the one preceding fertilization, (b) continue the present level, or (c) show no effect, that is, the level may continue to drop smoothly without interruption. Whichever manner the curve may develop is probably dependent upon the interval following last application, the amount of nitrogen previously absorbed, the amount of the current application, the age of the crop and the time of application with respect to season.

*Relationship Between Nitrogen Index and Growth:* The nitrogen curve itself was treated in the foregoing paragraphs. Its trend and development as influenced by fertilization were touched upon. The present discussion concerns the relationship between the nitrogen indices determined and the growth of sugar cane in pot culture.

It was shown that in the course of cane growth the nitrogen index of the plant fluctuated from 3 per cent as the maximum to 1 per cent as the minimum, and that the time required to reach the minimum was dependent upon the amount of nitrogen applied and absorbed, assuming that nitrogen alone was the limiting factor. The

problem, then, is to determine the significance of these values with respect to growth. Growth in this study refers to linear development.

*Reasons for Measuring Elongation:* It is admitted that elongation of the stem is only one phase in any growth measurement and that the gain in dry matter may proceed even when the linear development has slackened or ceased. However, until a method is devised which can integrate the various growth measurements, the estimation of elongation alone will have to suffice. Under ideal conditions where nitrogen is the limiting factor, it is generally accepted that a high supply of nitrogen in the plant is associated with increased or prolonged vegetative growth. Where such vegetative growth in sugar cane is allowed to proceed at an undiminished rate until close to harvest, it is believed that the resulting crop usually will be poor in quality although high in cane yield. Thus, for nitrogen fertilization, the optimum may be reached where vegetative growth can be balanced to permit greater opportunity for carbohydrate accumulation. Therefore, the measurement to be taken is one which will indicate vegetative growth or conditions for potential vegetative development. Since elongation is a function of vegetative growth, its measurement may then be taken for the purpose of this study.

*Correlations:* Growth measurements of the cane grown under the various treatments of this experiment are presented in Figs. 1, 2 and 3. The data cover the growth in the pots from the sixth to the thirty-sixth week. Correlation between growth and the nitrogen index is apparent from a study of these graphs.

It will be recalled that in this experiment the nitrogen for each treatment (amounts of nitrogen), except the  $1\frac{1}{2}$ -gram pots, was divided into two equal parts. The initial half was applied to all pots alike at two weeks after potting and the remaining half was applied either (a) when the first application was depleted, or (b) upon yellowing of foliage.

Examining Fig. 1 of the deficiency level, nitrogen treatment, and in particular the yellowing of foliage sub-treatment, it will be noted that the growth was affected by the amount of nitrogen applied and the time of application. The rate of elongation in the delayed application sub-series was higher during the sixth to the tenth week than between the tenth and sixteenth week. The total nitrogen content found in the leaf samples was between 3.4 per cent and 2.4 per cent in the first period and during the interval following was between 2.4 per cent and 1.3 per cent. When the final half of fertilizer was applied in the sixteenth week, the level was again raised to 1.5 per cent and growth increased. When the level dropped to about 1 per cent, the growth was practically negligible. In the complementary sub-series, when the second application was not delayed, growth occurred at a high rate between the sixth to the sixteenth week when the nitrogen indices were between 3.4 per cent to 1.5 per cent. When the level reached 1.3 per cent the rate of growth rapidly diminished, and upon reaching the 1 per cent level, growth was practically negligible.

As the nitrogen supply approached the sufficiency level, for instance in the  $1\frac{1}{2}$ -gram, single-application treatment, the growth curve paralleled the deficiency pots of 1-gram fertilization and continued to be correlated with the nitrogen percentage as in the lower treatment. This was shown in Fig. 3. However, as the nitrogen supply was increased with a higher level of fertility, the growth apparently did not correlate so closely with the nitrogen content of the leaf samples. Further examina-

tion at this stage will reveal interesting points on the relationship between nitrogen content of leaf specimens and growth of cane in pots.

It will be observed in the delayed-application sub-series of the 2-gram treatment, Fig. 2, that the growth occurring in the first application appeared to be still correlated with the nitrogen content during this period. However, after the second application, completing the sufficiency-level fertilization, growth did not follow the nitrogen curve as closely. This is to be expected since in the initial application the amount equalled only one gram of nitrogen, placing the supply still in the deficiency level so that the low level in the leaf nitrogen was reached early. In the other sub-series, when the last application followed the initial half immediately after soil depletion, growth was maintained at the initial high rate and did not slacken even when the 1.3 per cent level was reached for the leaf analysis. A closer study of the data will explain this apparent discrepancy between growth and leaf nitrogen content. Examining the upper portion of Fig. 4, it will be seen that the nitrogen content for the 2- and 3-gram treatments did not go below the 1.5 per cent level until the twenty-first and twenty-sixth week after potting and that upon passing this level the period of good growing weather was reached, that is, the summer months were approached. It therefore appears that good weather may act as a compensating factor for a low nitrogen level in the leaf area. In addition, translocation of nitrogen from other parts of the stalk may be sufficient to supply the needs for growth. Additional growth data for the entire period of the experiment are supplied in Figs. 5 and 6.

*Summarizing:* There appears to be a relationship between growth and the sufficiency of nitrogen and this sufficiency is indicated by the nitrogen content of the leaf sample. The critical low point below which growth stops is also affected by weather in addition to the sufficiency of the nitrogen supply.

From a study of the data at hand, it may be concluded tentatively that the nitrogen levels critically related to growth are:

2 per cent, above which there appears to be a luxury consumption.

1 per cent, minimum, desirable upon maturity.

Between 2 per cent and 1 per cent, growth proceeds normally, if other factors are not limiting. Above 1.5 per cent, nitrogen does not appear to be a limiting factor. Below 1.3 per cent growth diminishes or stops if weather is adverse. Growth may continue even at 1.3 per cent if weather is satisfactory. However, its rate may be affected.

If weather is optimum, the limiting level may be at 1 per cent, below which growth as elongation may be retarded. With good weather, translocation from other parts of the plant may furnish sufficient nitrogen for its growth, even if its level in the leaf area is at a minimum.

#### RESULTS OF FIELD SURVEY

Data and discussion are presented in the foregoing pages relative to leaf studies made with cane grown in pots. The question was raised as to whether data obtained from analysis of cane grown in the field under plantation practice are comparable to results similarly obtained from potted cane. In the early period of this investigation a survey was made in a number of plantation fields under the direction of Agee. Several plantations cooperated in making this survey. Samples were collected from random fields with the standardized leaf-punch procedure and analyzed

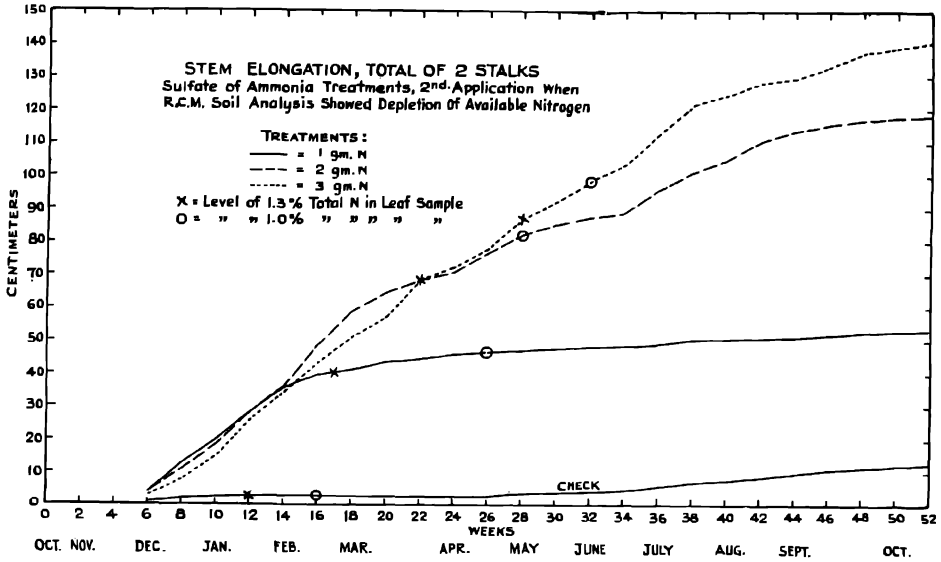


Fig. 5

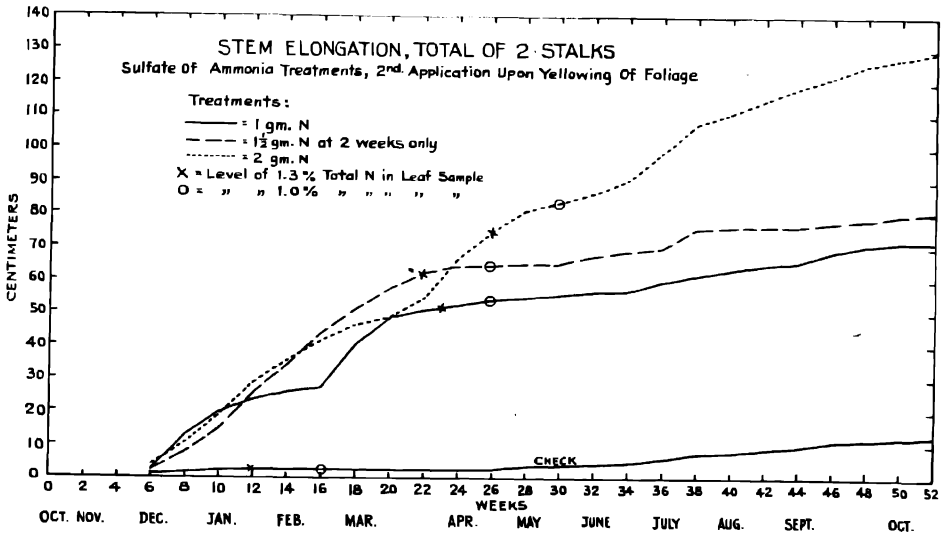


Fig. 6

by the regular rapid chemical method as previously explained. In the first step of this survey, collections were made from cane of different ages at one sampling period. Later the collection was extended so that certain fields were regularly sampled at progressive intervals. The objects of the survey were to determine the limits or range of nitrogen indices to be found in cane grown under regular systems of cultivation in established fields, to study the trend or the influence of age on the nitrogen content of the leaf zones believed sensitive and to learn facts regarding the variation in the nitrogen indices under the different conditions existing on various plantations. The results of this preliminary survey indicated that the range of values found in the field canes was similar to the one exhibited by the potted plants. The limits of indices remained between 2.7 per cent and 1.05 per cent. Young canes



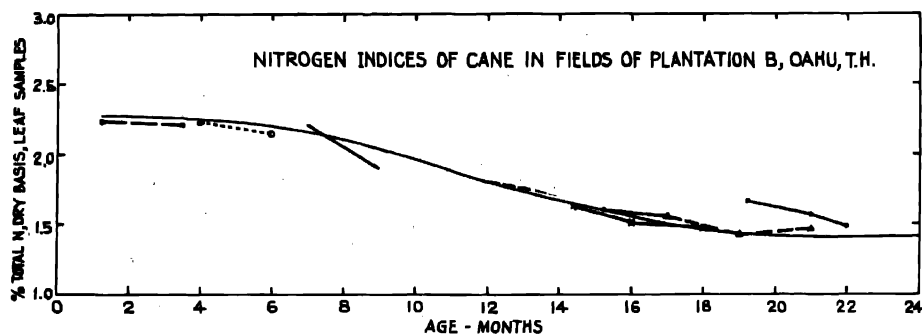


Fig. 7

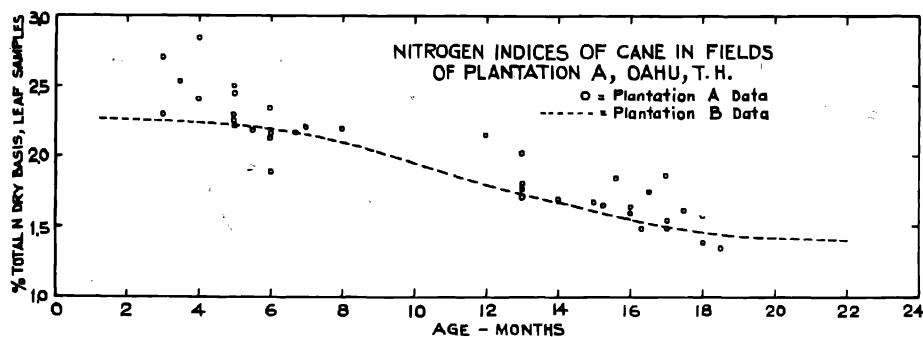


Fig. 8

up to about seven months of age were usually found to carry about 2.3 per cent nitrogen and those sampled at harvest, approximately 1.2 per cent for cane grown for about twenty-four months. Intermediate values were found for cane between the ages of 6 to 24 months. Part of the data from three plantations—two from Oahu and one from Maui—are presented in Table IX. It will be noted from these figures that the range of leaf nitrogen values were alike for a number of varieties and that there was a decrease in percentage content of the leaf zone progressively dropping with age of the cane.

As previously stated, a number of fields were sampled at regular intervals. In Fig. 7 are presented the data of Plantation "A," for eight fields, together with a smooth curve drawn by inspection, connecting these points. The rather uniform relationship existing between decrease of the nitrogen index with age of the cane grown on this plantation is illustrated by the curve. The variety under study was H 109. Fertilizer applied consisted of about 250 pounds nitrogen and applications were completed during the first six months of growth.

The data from Plantation "B," on this Island (Oahu), for H 109 cane are plotted in Fig. 8. The practice of the plantation for the crops under examination has been to apply approximately 250 pounds of nitrogen in two seasons, about two-thirds during the first season and the balance during the second season of growth. The smooth curve obtained for Plantation "A" is superimposed upon Fig. 8 so that a comparison may be made of the nitrogen indices obtained for cane grown in two different localities, reflecting probable differences in weather, fertilizer practices and soil and growth conditions. It will be noted that the points plotted for Plantation "B" in Fig. 8 are generally above the smooth curve representing Plantation "A" during the period under study.

TABLE IX  
ANALYSIS OF LEAF-PUNCH SAMPLES TAKEN IN SURVEY  
COLLECTIONS OF PLANTATION FIELDS

Plantation "A"—Oahu

Field	Variety	Age at sampling	N-index % total N
R3	H 109	1 mo.	2.33
G3	31-1389	4½ mo.	2.14
R10A	31-2510	5½ mo.	2.18
R10A	H 109	9 mo.	2.20
KL18	H 8965	9 mo.	2.32
G5A	H 109	12 mo.	1.99
R5B	H 109	14 mo.	1.82
KL19A	H 109	21½ mo.	1.46
R8A	H 109	23 mo.	1.32
H18	H 109	24 mo.	1.20

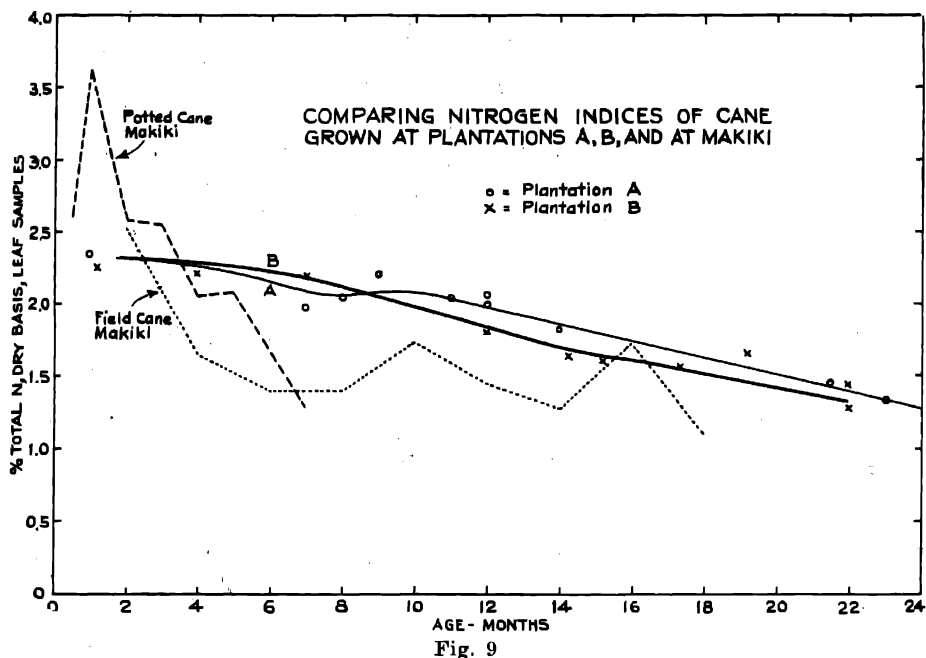
Plantation "B"—Oahu

Field	Variety	Age at sampling	Interval since last N appl'n	lb N applied	Total lb N applied	N-index % total N
13A	H 109	1¼ mo.	2 days	57	57	2.24
1A	H 109	4 mo.	2 mo.	151	206	2.22
20A	H 109	7 mo.	2 mo.	129	251	2.20
2A-3D	H 109	12 mo.	7 mo.	91	254	1.80
6	H 109	14¼ mo.	11 mo.	158	206	1.60
10C	H 109	17¼ mo.	12 mo.	106	250	1.56
21B	H 109	19 mo.	13 mo.	88	248	1.66
23B	H 109	22 mo.	17 mo.	101	254	1.42
23B—Edge	H 109	22 mo.	17 mo.	101	254	1.05
15A	H 109	22 mo.	15 mo.	77	238	1.28

Plantation "C"—Maui

39	POJ 2878	4 mo.	3 mo.	53	1.88
47 Mauka	POJ 36	4 mo.		0	1.64
47 Makai	POJ 36	4 mo.		0	1.84
48	31-1389	9 mo.	3 mo.	123	2.07
17	POJ 2878	10 mo.	4 mo.	98	1.85
43	POJ 2878	12 mo.	10 mo.	112	1.72
30A	31-1389	12 mo.	3 mo.	87	1.99
2	31-1389	13 mo.	5 mo.	109	1.45
21	Y.C.	18 mo.	9 mo.	158	1.40

The effect of climate and environment on the nitrogen content of leaf samples of cane grown in different localities is further illustrated in Fig. 9. Here a study is made of the field and pot data. The variety under examination is H 109 and the fertilization of nitrogen per acre basis is nearly alike for either field or pots. The Makiki field data originated from an experimental plot of cane grown at the Makiki station for cane composition studies by Cornelison and Ayres. The curves of Plantations "A" and "B" reflect samplings made during the spring of 1938, the pot data during the winter of 1937 and spring of 1938, and the Makiki field data from the summer of 1936 to the winter of 1937. The results, therefore, appear to indicate



that for a given variety of cane the curve of nitrogen percentages of the leaf samples collected at progressive intervals during the life of the crop may be affected by climatical and environmental factors (including soil and field practices). These factors affect growth and hence nitrogen absorption and utilization. These are reflected in the nitrogen indices determined.

#### *Relation Between Percentage Nitrogen Content of Leaf Samples and Growth of Sugar Cane Under Field Conditions:*

The similarity between the nitrogen content of leaves of potted cane and that of cane grown under field conditions as to nitrogen limits and the development of the nitrogen curve having been considered, the next matter of note refers to the relationship of nitrogen content of leaves and growth of cane in the field. Between the summers of 1936 and 1938, an experiment was conducted by Cornelison and Ayres (7) at the Makiki station in connection with the general research on basic studies of the sugar cane plant. During the period of the experiment, growth measurements of cane under various nitrogen treatments were taken by Cornelison. Leaf-punch analytical data were obtained by Mr. Davis for a study of the nitrogen economy of the sugar cane plant. In the present investigation, these data are re-examined and studied for the growth and leaf-punch nitrogen content relationship.

The elongation of primary stalks of the zero-nitrogen, the 100-pound, 250-pound and 400-pound nitrogen treatments, applied at the age of  $1\frac{1}{2}$  months, are shown in Fig. 10. The nitrogen percentage data of the corresponding treatments are also included in this chart. Growth measurements were taken from the fourth to the twenty-fourth month of growth while the nitrogen data covered only the period between the second and eighteenth months.

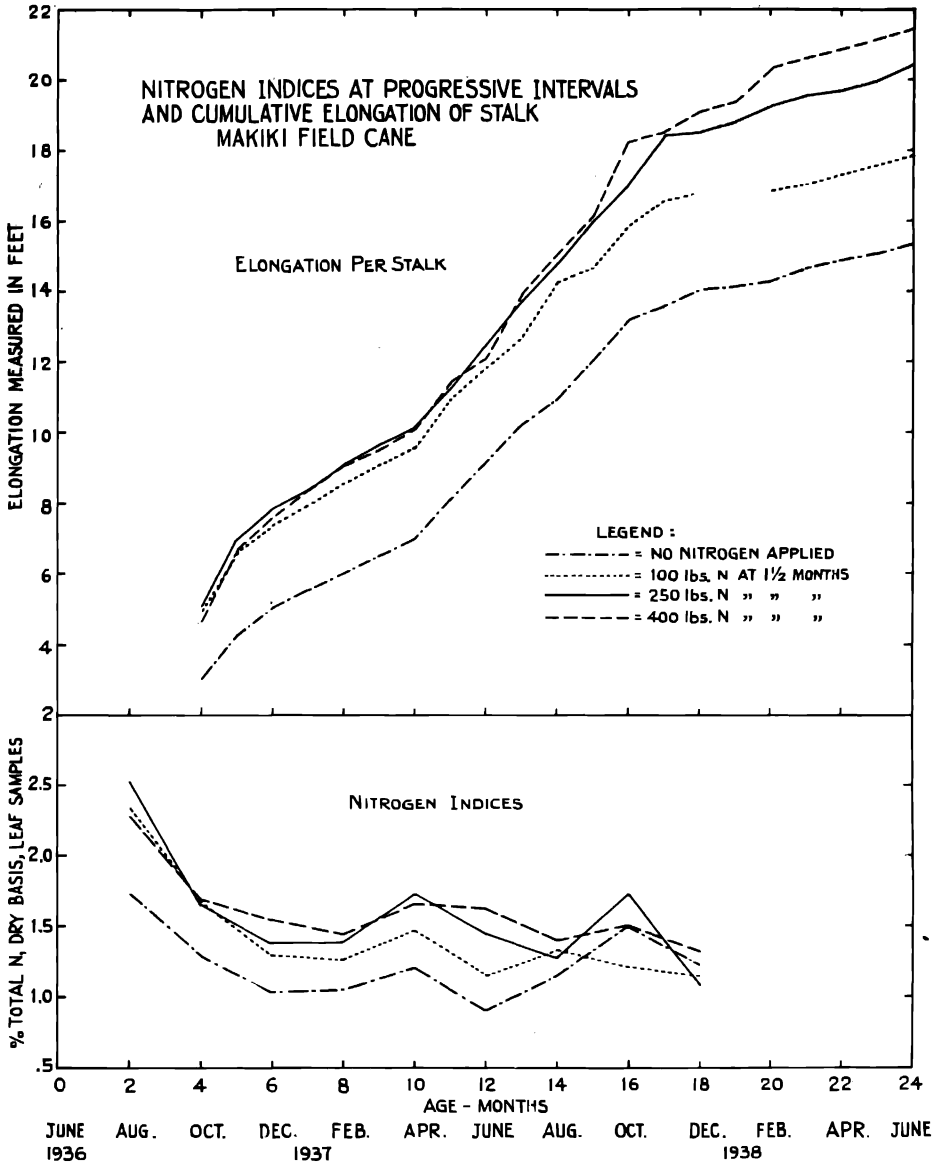


Fig. 10

Referring to the nitrogen curves of Fig. 10, a correlation appears to exist between the nitrogen levels and the fertilization made. In the zero-nitrogen plots, the level dropped from a high of 1.7 per cent to a low of 1.0 per cent within the interval of four months (age two to six months). In the treated plots, the levels were nearly alike for all treatments from the second to the fourth month of growth. From the sixth to the twelfth month a correlation appears to exist for the different amounts applied. During the first year's growth, the percentage nitrogen level of the leaf samples was lowest for the zero plots, while the 100-pound treatment was higher and did not drop to the level of the zero-nitrogen treatment at any time during this

period. It ranged from 2.3 per cent to 1.2 per cent. The next higher level was shown by the 250-pound treated plots. The highest level was reached by the 400-pound plots. However, the levels for both the 250- and 400-pound treated plots were nearly alike.

Turning now to the elongation curves, it will be seen that the total elongation appeared to be correlated with the fertilization. In the zero-nitrogen plots, growth was lowest, the next higher was the 100-pound treatment followed, still higher, by the 250- and 400-pound treatments, these latter showing very much the same elongation. Considering the elongation from the standpoint of nitrogen levels at any moment, it appears that even in the no-nitrogen plots elongation did not cease even when the level of 1.0 per cent was reached at the age of six months. As discussed in the pot studies, growth does not always stop when this low level is reached if other favorable factors for growth, especially weather, are optimum. It appears from the graph that the low level did not remain fixed at 1 per cent, but fluctuated occasionally above this value. It was shown in the other studies of this experiment by Ayres that the total nitrogen content of the entire aerial portion of the plant continued to increase even after the twelfth month, indicating, perhaps, that a small amount of soil nitrogen was continually becoming available and then absorbed. However, in spite of the continued growth, it appeared definitely from the study that with a low percentage nitrogen level, growth did not equal the fertilized pots at the start nor did it catch up with the others. In the 100-pound treatment, the low level fluctuated in the vicinity of 1.3 per cent which, as previously stated, was just about the lowest level to which the nitrogen value may drop without impairing growth. For the higher applications, the low level fluctuated closely around 1.5 per cent and the growth for these treatments was highest.

From results of this study of field data, the conclusion may be drawn that nitrogen levels in relation to stem elongation are effective for cane growing under field conditions as well as when cultivated under potted environment. It appears that if the nitrogen index level is at 1.5 per cent, H 109 may continue to elongate at a high rate if other factors are not limiting.

Having determined the development and trend of the nitrogen percentage curve of the leaf system of a growing cane crop and established the relationship between nitrogen levels in the leaf specimens and growth (elongation) of sugar cane, the matter of practical applications will be considered.

#### DEVELOPING TENTATIVE PROCEDURES FOR NITROGEN CONTROL

##### *A Review of Other Experiments:*

Considerable attention has been devoted by many investigators in recent years to the development of methods of plant analysis which may lead to a determination of the mineral requirements of plants. Either the entire plant or parts thereof have been analyzed in seeking a means of learning its nutrient requirement. Leaf analysis as a proposed measure has the support of results obtained in many such studies. Thomas (14, 15) has described a system of leaf analysis based on the results of his own experiments on potatoes and apples considered with those of other continental investigators. This system of determining plant nutrient requirement is

known as "foliar diagnosis" and is defined by Thomas as a measure of the chemical condition or state of leaf with respect to the dominant nutritive elements at the instant of sampling. The sample is taken from a pre-determined and suitable position on the plant stalk. The foliar diagnosis for any given interval is defined as the sequence of chemical composition as determined at different periods during the growth period. This method departs from the usual in obtaining an analytical expression of plant performance in that the chemical composition of the leaf at different times is measured rather than determining the nutrient status of the soil or considering the nutrients added.

As previously stated, Lundegårdh (11) has proposed the analysis of the leaf in conjunction with soil analysis to determine the need of the plant *and* the fertility of the soil. According to Lundegårdh, growth and development of the plants are primarily dependent upon the inner concentration and distribution of the nutritive elements. From results of his study with oats he suggests that the ash of the leaves, within certain limits, will reflect the nutritive condition of the whole plant. He concludes that growth will cease when the concentration of any of a number of elements (especially potassium, calcium, phosphorus, and probably manganese and iron) in the leaves falls below a certain minimum.

The object of plant analysis, in addition to determining the nutrient requirement of a crop, has been in many instances based on a determination of the supplying power of the soil for available nutrients. Since it has often been found that the soil does to a large extent influence the composition of a plant growing upon it, the attempts to evaluate soil fertility with plant analysis does not seem entirely illogical. However, results obtained by many investigators have indicated that the nutrient content of a plant does not always necessarily reflect the supplying power of the soil. Thus, a soil deficiency of one element may result in a high level of another element appearing in a plant grown on that soil. This may not, however, indicate a high level of fertility of the latter element in that soil. Again, a low nutrient level may exist in the plant which may not correlate with a corresponding low, soil nutrient availability. Nitrogen is one element which may behave with such characteristic inconsistency. This fact has again indicated the necessity of placing greater emphasis on looking to plant analysis as a means of determining the nutrient requirement rather than as a medium of estimating soil fertility at least for nitrogen.

Macy (12) apparently has demonstrated the successful application of plant analysis to this phase of the nutrient requirement problem of plants. From his studies of the results of a number of investigators on plant growth, fertilization and nutrient content of plant materials produced, and from his own experiments with barley, Macy has advanced a theory on the relationship existing between the percentage content of a nutrient in a plant and the sufficiency of the nutrient for growth. This theory has been proposed for application in a method to determine the mineral nutrient requirement of plants by plant analysis. The dominant concept of this theory, as proposed by Macy, is a critical percentage of each nutrient in each kind of plant, above which there is *luxury consumption* and below which there is poverty adjustment which proceeds until a minimum level is reached.

During the period of poverty adjustment, growth takes place according to Mitscherlich's law of minimum and a factor may be only partially limiting because

of the compensating effect of other factors which may prove more favorable. Above the critical percentage (luxury consumption) or during the period of minimum level, Liebig's law of growth is in effect, which holds that the yield is directly proportional to the supply of the nutrient. While admitting that the percentage content of a nutrient in the plant may be affected by other growth factors, Macy points out that under the circumstances the sufficiency of the nutrient, as measured by the response to it, is likewise affected so that the relationship still holds. However, it was indicated that the critical and minimum percentages of nutrients in plants are not rigidly set, but may fluctuate because of the association of elements; one may reduce the effectiveness of another or may partly substitute for another. For instance, it was pointed out that the critical percentage of phosphorus may be raised by the presence of larger than normal quantities of aluminum in the plant.

In order to test the theory of sufficiency in fertilization, Macy conducted pot experiments growing barley in different soils and using fertilizer treatments with varying increments of nitrogen. Yield data for different increments of nitrogen were obtained and the plant materials produced were analyzed for total nitrogen. The yield data showed very large effects of treatments and of soils. While a wide variation occurred in the absolute yields, the relationship between the percentage nitrogen content of the straw and its response to nitrogen was independent of other variable factors. Thus, while the yield data for one treatment were definitely different from another, the points indicating the relationship between per cent nitrogen content and the straw of barley produced fell within a narrow curve and did not vary in excess of twice the probable error for a single point. The critical percentage determined by Macy for barley was 0.8 per cent nitrogen, and the minimum percentage about 0.4 per cent. Additional data were cited on the percentages for many plants determined by others which were comparable to Macy's critical percentage. These data follow: Wolff found 1 per cent nitrogen for the oat plant and 0.35 per cent  $P_2O_5$ ; similar nitrogen levels for oats were apparent from results of Pfeffer, et al; Fraps, studying corn grown in pots, reported that 82 per cent of the crops having more than 0.70 per cent nitrogen responded to N less than 10 grams per pot, while 82 per cent of the crops having less than 0.70 per cent nitrogen responded to nitrogen more than 10 grams per pot; the sulfur content of alfalfa was indicated by Alway to be 0.2 per cent S; Garner obtained preliminary data indicating the same value for sulfur in tobacco; and Garner, et al, reported a tentative critical percentage of 1.5 per cent N in tobacco leaves.

#### *Development of the Procedure:*

Having considered briefly the theories advanced by other workers to determine the nutrient requirement of plants by analysis of their tissues, it is proposed to ascertain whether they are applicable to the method of leaf-punch analysis in controlling nitrogen fertilization of sugar cane. From a preliminary study it appears that the concept of critical and minimum percentages may be applied to leaf-punch analyses performed over intervals during the growth of the crop and a practical method of control may thus be formulated.

The data for yield and nitrogen content in Macy's calculations are obtained upon maturity of the crop grown with different increments of nitrogen. In the proposed R.C.M. leaf-punch procedure, the nitrogen values are obtained at pro-

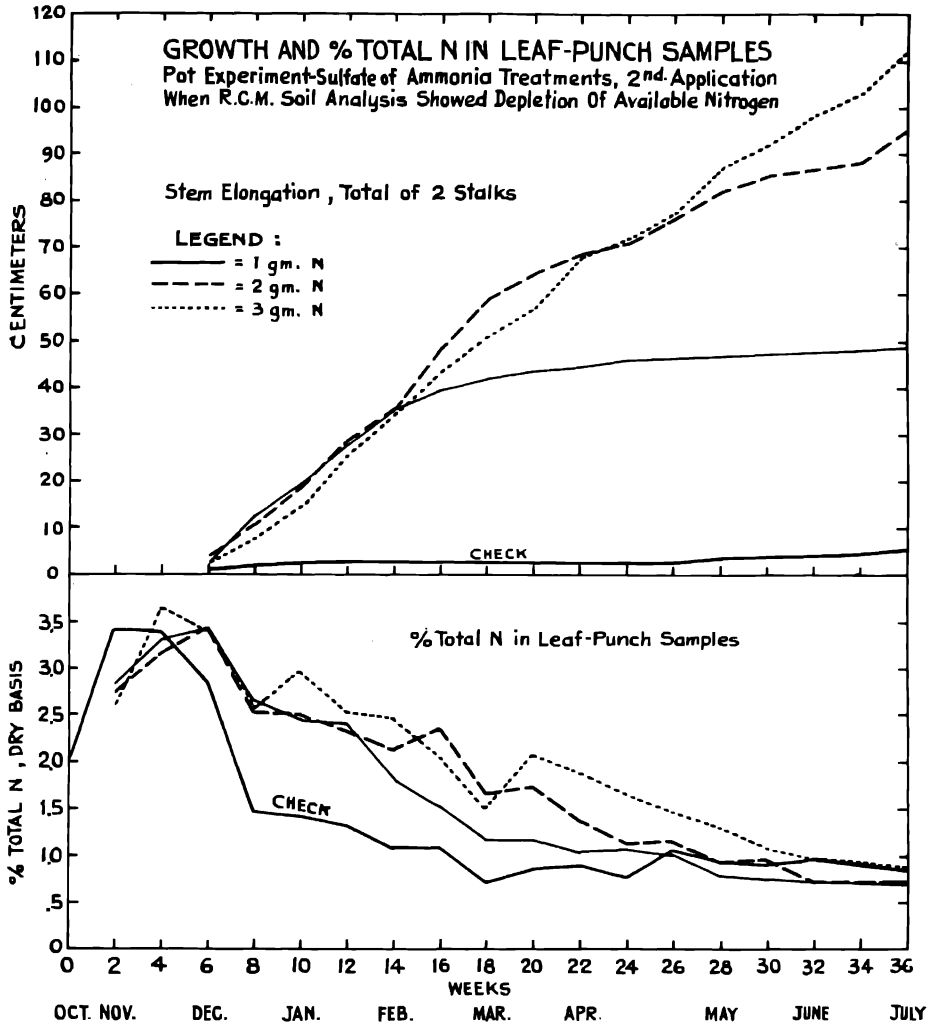


Fig. 11

gressive periods of growth. Thus, while Macy's data are obtained for a single period, the method herein proposed furnishes data for a successive number of periods. The interpretations placed upon both systems as to sufficiency, however, are comparable since their derivation comes from a common basis, that is, based upon the response of the plant to the fertilizer applied.

Reviewing briefly the relationship between nitrogen content of sugar cane leaves and the growth of cane as previously discussed, it may be restated that apparently, at least for H 109 cane, there is such a positive correlation. The data for pot culture of H 109 fertilized with sulfate of ammonia are summarized graphically in Fig. 11. The levels tentatively established from the study are, critical percentage 2.0 per cent, minimum percentage 1.0 per cent, and poverty adjustment between 2 and 1 per cent.

The object of the control is to maintain a balance in growth so that the excessive vegetative activity may not be too prolonged during the development of



sugar cane from start to harvest. Vigorous vegetative activity is generally believed to favor sugar consumption rather than sugar accumulation. Its regulation, therefore, appears to be the desirable approach in determining the proper nitrogen requirement of the crop.

Trends in the rate of elongation of stalks are apparent from a study of the growth curves for a number of sugar cane crops grown under Hawaiian conditions. Weather and sufficiency of nutrient supply considered, it may be reasonably expected that in general the rates apparently characterize three stages of growth which become readily recognized. First, during the initial development of the crop, growth occurs slowly. Fertilizers are applied during this period. The nitrogen content of the foliage at this stage becomes rather high. With fuller development of the plant, the rate of growth gradually increases until a period occurs in which the rate of elongation has increased to a maximum and growth is rapid. This is the step which is generally recognized as the "boom stage" of growth for sugar cane. Usually, as expected under the established general fertilization practice, first-season nitrogen applications will have been completed within the early part of this boom stage of development. Sometime within this period a point will be reached where the rate of growth will be increased relatively greater than the rate of nitrogen absorption so that as elaboration proceeds the percentage of nitrogen in the plant will begin to decrease. As the nitrogen supply diminishes and as the meteorological factors become limiting, the rate of growth will gradually become retarded so that in the final stage of the crop cycle elongation gradually slackens and proceeds slowly or may cease entirely.

In the earlier discussions it was indicated that nitrogen fertilization may affect the percentage content of this nutrient in the sugar cane leaf and there is an apparent relationship between the percentages determined and the development of the plant. The proposed fertilization control, therefore, may be maintained, it seems, by regulating the nitrogen level throughout the cycle of crop growth. The initial development of the crop is planned to start and continue with a nitrogen level in the leaf close to the critical percentage. The growth at boom stage and throughout the period of accelerated elongation is to occur concurrently with the gradual lowering of the nitrogen percentage which will be in line with the poverty adjustment discussed previously. The final ripening will then take place as the minimum level of this nutrient is reached. The usual crop length under Hawaiian practice is about twenty-four months from start to harvest. The problem then is to space this interval so that a sufficient period is given to develop a good start, followed with an interval of vigorous vegetative growth and ending with a progressively decreasing rate of elongation and development in order that sufficient time for ripening may be allowed.

From a study of numerous field leaf nitrogen data, the results appear to indicate that the levels established below may meet the requirement of a 24-month crop:

Level	At age
2.0%	6-8 months
1.5%	12-14 months
1.25%	15-17 months
1.1%	24 months

Considering the usual practice of applying nitrogen fertilization in quantities sufficient for a 24-month crop, the results at hand indicate that during the initial period of growth it may be necessary to maintain the nitrogen level at about the critical percentage (2 per cent) for about six months from the start of the crop. This is to insure the proper development of the plant, that is, apparently nitrogen in excess of current requirement is required in order that tillering may occur early. If only a minimum supply of nitrogen is present, it may be possible that only the primary stalks will grow without much development of suckers. Hence, under a minimum supply of nitrogen, the quantity of growth upon maturity may be insufficient. Therefore, the critical level perhaps should be maintained for a period of about six months. Since it has been indicated that the level above 2 per cent is approaching the luxury consumption status, the level does not appear to be required, at such times, much above 2 per cent, except that during the first two or three months of growth following planting or ratooning the level may be higher than 2 per cent and it usually is (approximately 2.5 per cent, more or less) for this stage of growth.

After this initial development at about the critical limit, it is believed that the percentage should drop, commencing with and during the boom stage of growth. At this period "poverty adjustment" should set in. It has been shown previously that the interval in which the poverty adjustment operates, that is, the drop from the critical to the minimum percentage, will depend upon the nitrogen supply and the assimilation of nitrogen resulting from development and growth. Hence, if the nitrogen supply is excessive, the interval of the critical percentage will be maintained for a period much longer than six months and the time required to complete the poverty adjustment will be prolonged. The results of this study appear to indicate that if the nitrogen supply is not excessive growth should proceed so that the level drops gradually after the initial period from 2 per cent to 1.25 per cent upon reaching ages of 15 to 17 months. It has been shown in the growth studies previously described that elongation will not be retarded if the nitrogen level drops to 1.5 per cent or even as low as 1.3 per cent. If the level is maintained at 1.5 per cent, or above, for a considerably longer period than was indicated (a result, apparently, of the crop being over-fertilized, as has been shown to be possible) the conditions for vegetative growth are then maintained above normal for the corresponding age of cane. This may result in delayed or insufficient ripening before the 24-month harvest.

Further growth after the 15- to 17-month period may deplete the nitrogen supply in the plant so that the level will continue to drop. This should enable the plant to undergo a favorable and sufficient period of ripening which would favor its reaching the minimum N level of about 1 per cent at 24 months.

A form has been developed for recording the leaf-punch nitrogen data obtained in this type of study. It is illustrated in Fig. 12. Spaces are provided for data pertinent to field, crop and fertilization. Tabular columns are included for recording date of sampling, age of cane at sampling, growth measurement data if taken, the nitrogen index values and remarks. At the bottom of the page a form is appended for plotting a graphical presentation of the nitrogen data. The nitrogen percentages are placed on the ordinate and the age in months on the abscissa. In this graph, emphasized by heavy rulings, are shown the limits discussed above so



that the trend and concentration of the nitrogen supply may be observed from the graph at a glance.

The limits of nitrogen variation in cane leaves for a 24-month crop have been discussed. However, some crops are grown for shorter periods, usually between 14 to 18 months. Since the growing period is thus shortened, and in order to provide a period of sufficient duration for ripening, the program of fertilization should be modified. However, in order to insure a proper development of the stool at the start of the crop, it may be found expedient to decrease the amount of nitrogen applied only to such an extent which will still insure a maintenance of the critical percentage up to the ages between 4 and 6 months. Sufficient data of this order have not been accumulated to justify a recommendation of fertilization establishing even tentative limits for the short crop. However, as a suggestive basis and in lieu of further study of the needs for this type of crop, limits of nitrogen levels which may be found appropriate are offered below :

N level	At age of cane
2.0%	4-6 months
1.5%	8-10 months
1.25%	10 months, lower limit
1.25%	18 months, upper limit

The method to control nitrogen fertilization of sugar cane by analysis of leaf-punch samples will now be summarized briefly. The system consists of a plant nitrogen study, followed through from start to harvest. Leaf-punch samples are taken at regular and progressive intervals and analyzed for total nitrogen. First sampling is usually started with 3-month cane and continues at about monthly intervals up to harvest. The results obtained are designated as the "nitrogen index" of the plant at a given age. These indices do not represent the exact quantity of nitrogen in the general leaf system or the entire plant but serve only, it is believed, as indicators of plant performance. Their interpretation with respect to nitrogen index limits proposed for cane of different ages may serve to indicate whether the nitrogen supply is sufficient, deficient, or in excess at the moment of sampling. The analytical data are recorded numerically and graphically. In the form of a graph, the points representing nitrogen index at progressive periods of growth form a curve upon which are superimposed the limits of the significant nitrogen levels previously discussed. The crop nitrogen situation, if plotted on the prescribed graph, is therefore available to the observer in a rather simplified and yet comprehensive form.

#### PRACTICAL APPLICATIONS

##### *Application With Respect to Preharvest Samples:*

While studied primarily for fertilization control of a crop by following it from start to harvest, the method may offer possibility of obtaining further information of value in preharvest sampling as it is now conducted by several plantations. A tabulation of data from regularly harvested plantation fields is presented

TABLE X  
ANALYSIS OF LEAF-PUNCH SAMPLES OBTAINED FROM CANE PRIOR TO HARVEST AND AT HARVEST.  
FIELD, ANALYTICAL AND YIELD DATA ARE TABULATED.

Field data			Lbs. N applied per acre	Nitrogen indices of samples taken, months before harvest				Yield data			
Plantation	Field	Variety		3	2	1	At harvest	Age at harvest	TCPA	TSPA	TC/TS*
"A"	K7B	H 109	230	1.75	1.68	1.60	1.50	19½	83.60	9.72	8.60
"A"	O-1B	H 109	236	1.58	1.56	1.57	1.55	21	83.83	9.52	8.81
"A"	O-7	H 109	238	1.35	1.48	1.41	1.28	22	89.82	10.81	8.31
"A"	G11	H 109	217			1.55	1.39	18½	71.16	8.45	8.43
"A"	P-1	H 109				1.49		18	71.74	7.57	9.47
"A"	O-16	27-8101	240				1.72	16½	94.63	9.61	9.85

Field data			Lbs. N applied per acre	Nitrogen indices of samples taken, months before harvest				Yield data			
Plantation	Field	Variety		3	2	1	At harvest	Age at harvest	TCPA	TSPA	QR*
"B"	30B	H 109	253		1.56	1.43	1.26	22	62.85	7.20	9.13
"B"	2A-3D	H 109	254	1.71			1.42	17	89.95	10.23	9.78
"B"	6	H 109	206		1.51		1.48	18	92.93	10.74	9.21
"B"	10C	H 109	250		1.42		1.47	21	89.62	9.70	10.04
"B"	21B	H 109	248	1.66		1.56	1.48	22	75.33	8.60	9.94
"B"	15A	H 109	237				1.28	22	105.76	12.74	8.30

\* For Plantation "A"—Tons cane per ton sugar (TC/TS) are reported in place of the quality ratio (QR).  
For Plantation "B"—The regular quality ratio (QR) data are presented.

in Table X. The relatively few cases of this type which have been studied do not warrant the assumption of conclusions which are necessarily significant. However, the generally poor juice qualities noted for some crops may appear to be correlated with the higher nitrogen percentages found in the corresponding leaf samples collected at harvest. The leaf-punch nitrogen results obtained at harvest having indices of about 1.5 per cent may be considered high insofar as they indicate a relatively ample sufficiency of nitrogen available to the crop to support further vegetative growth. The relationship between ripeness and quality of cane is not a subject the authors are qualified to include within the scope of this paper. The data are presented as such for suggested further study. It may be found that if preharvest data are to be rigidly interpreted, a single nitrogen value taken only at the time of harvest may not be found as valuable as a number of nitrogen indices determined at regular intervals prior to harvest. Preharvest sampling so far as leaf nitrogen studies are concerned should preferably commence just prior to the cessation of irrigation and continue at intervals through ripening until the time of harvesting.

*Application in Control of Fertilization—A Tentative Procedure:*

A few cases will be cited to illustrate possibilities of the practical application of the method in nitrogen control. Certain conclusions drawn from the experimental work of this study and a few accepted generalizations resulting from practical experiences of many local workers in sugar cane culture will be presented preparatory to the discussion on applications. First, it may be stated that the method here proposed does not directly control the quality of the cane produced. The type of study and the results obtained do not warrant any conclusions as to the relation of nitrogen percentages or growth to quality. Their relationship, if apparent in this study, has been noted only as an indirect observation. This method is designed for controlling the nitrogen *levels* in the cane leaf system because the variability of these levels appears to be associated with growth, that is, elongation of the stalk or the persistence of vegetative activity. The levels suggest a relationship bearing upon the sufficiency of the growth factor. In this study it has been assumed that the nutrients other than nitrogen have been supplied to or are present in the soil in amounts sufficient to insure that nitrogen only is the limiting nutrient. Second, it has been found that most Hawaiian soils planted to sugar cane will respond to nitrogen fertilization. A lack of response has been reported in only a few exceptional instances. (The amount of nitrogen required per crop may, however, vary with the variety from crop to crop and from locality to locality. In general, response has been realized up to applications of 150 pounds of nitrogen per acre, but very little economic response is reported where the application equals or exceeds 300 pounds nitrogen per acre.)

Assuming that a field of cane is to be fertilized and that the previous crop did not show a substantial gain when receiving over 150 pounds of nitrogen, then, for the first application an amount may be applied which will be *less* than the 150 pounds required for the previous crop. The quantity needed is split into a number of applications, or the entire amount is applied initially, depending upon the practice in vogue at the plantation. Commencing with the age of three months, or earlier, if the development of the shoots permit earlier sampling, the first leaf-punch

samples are collected and this practice continued periodically thereafter at regular intervals. The results obtained are recorded numerically and graphically on the forms provided for that purpose. The data determined at each sampling with reference to its index of sufficiency as compared with the theoretical curve will suggest whether additional nitrogen may or may not be applied. The quantity of nitrogen to be applied will depend upon (a) previous requirement of the crop, (b) the age of the present crop, (c) the nutrient level as to sufficiency, (d) the time of the year, (e) the total quantity applied to date, and (f) the final total which will be reached with the addition of the current increment. In the un-irrigated plantation, it may be of advantage also to determine available soil nitrogen. The above brief discussion illustrates, in general, the procedure which may be followed in applying leaf-punch nitrogen analyses to nitrogen fertilization control. Until a much wider experience is gained in the field, the suggested procedure of study can scarcely be made more specific.

A hypothetical case is presented to demonstrate the practical application of this system. Referring to Fig. 10, data have been plotted from the results of the experiment where 250 pounds of nitrogen were added in a single application to a plot of cane at the age of  $1\frac{1}{2}$  months. The crop was planted in June. The analytical data indicated that at the age of 3 months the nitrogen level was down to 2 per cent (by inspection) and at the age of 4 months it had dropped to 1.7 per cent. Considering the theoretical nitrogen limits for the cane, the percentage of this nutrient determined indicated an apparent need for nitrogen at this point. It now becomes a question as to whether more nitrogen should or should not be applied. A suggested first step would be to resample the cane leaves to learn if the value of 1.7 per cent was correct. Having checked this finding, the soil may be analyzed to determine the supply of nitrogen available to the crop. Assuming that analysis showed depletion of available soil nitrogen and that the stand of cane appeared normal, vigorous and well developed, the investigator may then conclude that in view of the low nitrogen level in this young cane, assuming that fair growing weather prevails, further fertilization is in order. The decision having been made to apply nitrogen, it becomes necessary to decide on the quantity to add. From past experience one may realize that cane in this general area did not respond to nitrogen applications above 250 pounds. Knowing that in general this amount represents nearly the upper limit of applied nitrogen with most cane areas, it may therefore appear as unwise to increase the application by 60 or 75 pounds more and thereby bring the total applied in excess of 300 pounds. Therefore, let it be assumed that 50 pounds additional nitrogen are applied. If the subsequent cane leaf analysis two months later shows a still lower level, say 1.4 per cent at 6 months, the question of additional fertilization will again have to be considered.

Based upon the interpretation of the data alone as to sufficiency for a 6 months' crop, the leaf nitrogen at 1.4 per cent will appear to indicate a need for this nutrient. However, other factors will have to be considered, as pointed out previously. One of them is the final total quantity which will be reached if further increments are to be added. Since the quantity of nitrogen added to date has totalled 300 pounds and the optimum for previous crops was believed to be 250 pounds, any additional application will require careful consideration. It was previously discussed in the relationship of nitrogen levels to growth that 1 per cent is the mini-

imum level in which growth will be retarded. Also, it was indicated that at 1.3 per cent growth will not be limited if weather is optimum. The level of 1.4 per cent, therefore, while unusually low as far as being best for the initial development of the crop, need not be considered as absolutely limiting. The cane is 6 months old in December. Hence, growing weather for the following two or three months may be assumed in perspective as being not optimum, based upon the usual winter seasonal behavior. All factors considered, therefore, the decision may rightly be made to leave off fertilization for the next two months.

The level at 8 months still shows a nitrogen index of 1.4 per cent. If a deficiency had then existed, growth and elaboration during the previous two months should have lowered the level. Since the level did not drop in this interim but remained at 1.4 per cent, it seems in order at 8 months to still delay further application for another two months of cold weather. After this interval it is expected that better growing conditions will have set in. Fertilization made at that time will be in line with the customary second-season practice and is therefore not too late. However, from the tenth month on, the level rose and followed the theoretical limits closely up to the age of eighteen months, when further leaf collections were discontinued. Results of the actual experiment support the decisions made in the hypothetical case. Treatments of 100 pounds and 400 pounds were made in addition to one supplying 250 pounds nitrogen in the actual test. The growth data (both elongation and increase in dry matter per stalk) seem to indicate that the 250-pound application was the optimum. The 300 pounds made in theoretical applications to the hypothetical case appear sufficient and yet not excessive. The quantity is practically close to the optimum.

Consider next an actual case illustrated in Fig. 13. In this field of H 109 cane the initial amount of 50 pounds was applied to a plant crop at the age of  $1\frac{1}{2}$  months. Leaf-punch analyses within an interval of 2 months after fertilization showed the nitrogen level to have dropped from a high of 2.4 per cent to 1.95 per cent. An additional 50 pounds were applied. Subsequent analyses showed a drop to 1.74 per cent at the age of 5 months, indicating a necessity for further fertilization. Fifty pounds more were applied. This addition raised the level to 2.07 per cent at the age of  $6\frac{1}{2}$  months and two weeks later dropped to 1.92 per cent. According to the proposed limits further fertilization for the time being was not necessary, but a contingency in the cultural practice for this field required an additional 75 pounds. Samples taken for the next two months indicated that the additional 75 pounds had created an excess. The requirement for this particular crop, according to interpretation of analytical data, apparently should be between 150 to 175 pounds.

In the other half of the same chart is illustrated the requirement for POJ 2878 cane grown in the same field and under identical conditions. The nitrogen curve for this variety indicated that the 225 pounds were sufficient for the crop up to the ninth month. Up to this period the curve, while slightly low for the first period of growth, followed closely the theoretical limits. The points on this curve, while below 2 per cent, were still higher than 1.5 per cent. Since 1.5 per cent is not deficient for normal growth, it may be assumed that the curve is satisfactory. The curve continued to drop below the arbitrary limits at the ages of 10 and 11 months, the nitrogen index at 11 months being slightly below the 1.5 per cent



## NITROGEN INDICES DETERMINED DURING PROGRESSIVE INTERVALS OF GROWTH

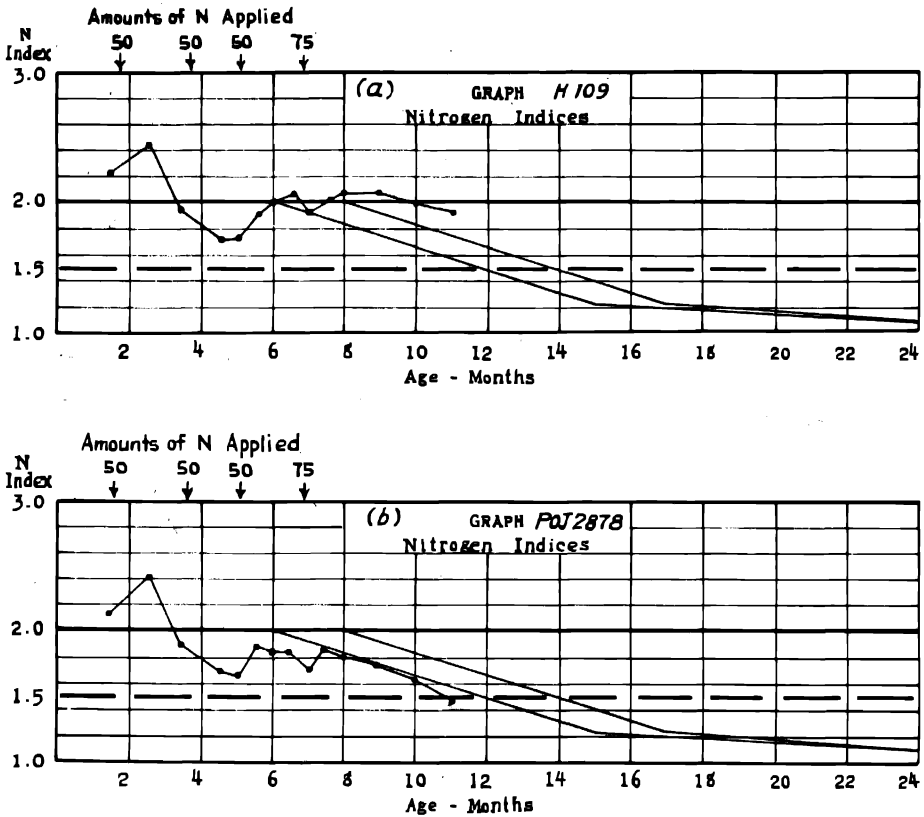


Fig. 13

level. According to the proposed limits, second-season applications of nitrogen may be made if the crop is to be grown for a 24-month period. Additional field data illustrating trends obtained in practical application are presented in Fig. 14.

It is to be hoped that with this method there will be placed in the agriculturist's hands an additional instrument which may aid him in putting nitrogen fertilization upon a more nearly rational basis of field control. During the course of this investigation a large number of Grade "A" amounts-of-nitrogen experiments were studied. The results appear to indicate that in many of these experiments the field practice applications exceeded the optimum requirement by as much as 50 to 100 pounds of nitrogen—equivalent to 250 to 500 pounds of sulfate of ammonia fertilizer. These excesses were in many instances not only for one crop, but were shown to exist in the preceding one or two crops. In spite of this, for lack of a means or method which will evaluate the variable requirement of a crop, the large applications rather than the indicated optimum have been maintained more or less as precautionary measures. With the tentative procedure outlined, it is possible to obtain from a growing crop data which may be measured against tentative standards to determine the assimilation of nitrogen or its relative sufficiency. These data will offer an additional guide which the agriculturist may consider in arriving at a decision on crop nitrogen requirements.

# NITROGEN INDICES DETERMINED DURING PROGRESSIVE INTERVALS OF GROWTH

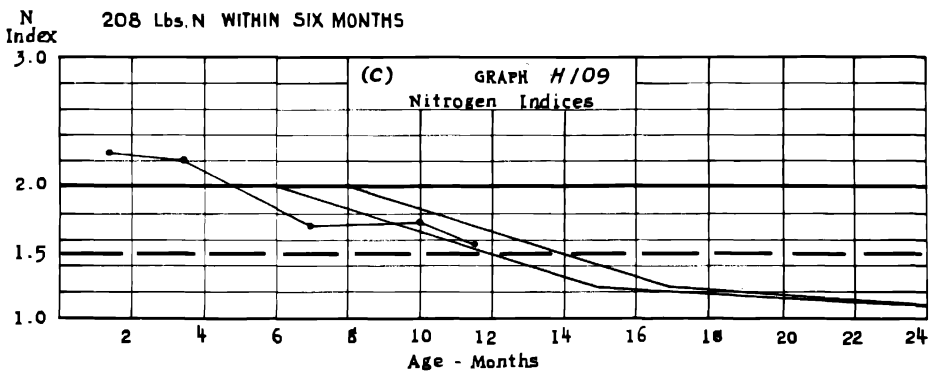
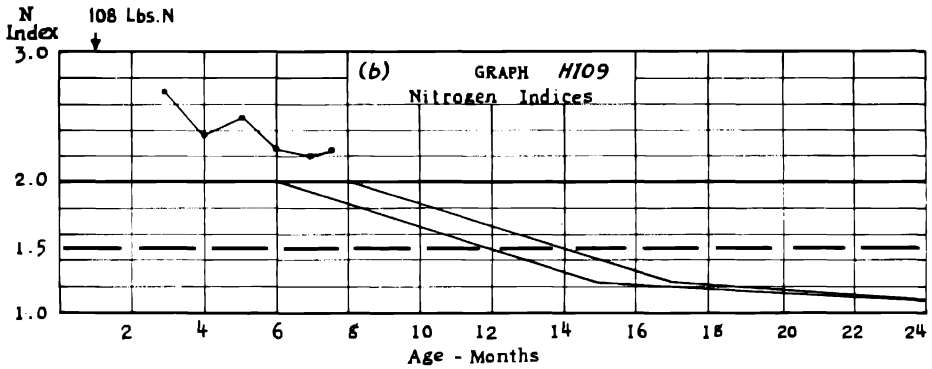
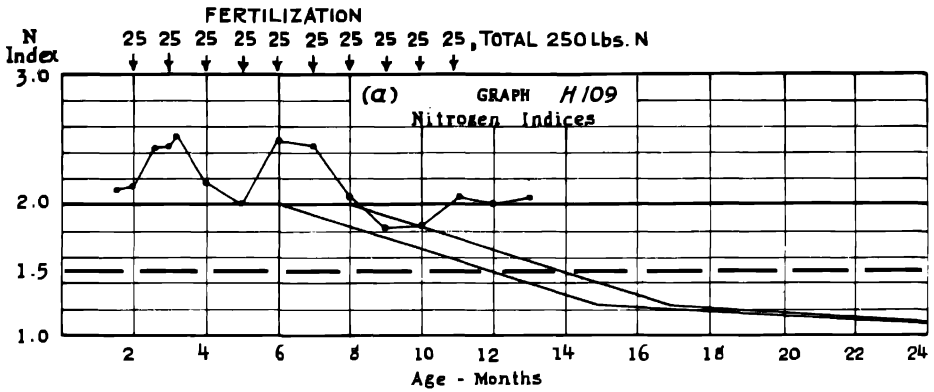


Fig. 14

## SUMMARY

Sugar yields in Hawaiian production in recent years have indicated the necessity for closer control of nitrogen fertilization. The determination of *available* nitrogen by soil analysis has been found to possess only limited application in crop fertilization control of nitrogen. Complicating the problem is the changing requirement of the crop for nitrogen as influenced by weather. These factors have pointed to a desirability of learning from the plant itself its needs for nitrogen.

The possibility of using the plant leaf system as an index of its nitrogen needs was explored with a rapid chemical method developed at this Experiment Station. A standardized procedure for obtaining leaf-punch samples from definitely localized points in the leaf system for nitrogen analysis was established through the auspices and active participation of Mr. Agee. The results indicated that the leaves designated by the second, third and fourth dewlap (ligule) attachments gave the most consistently satisfactory results. The uppermost visible dewlap is counted as one. The particular section of the leaf to be sampled has been found to exist in a region midway of the blade from either end at its widest point. The procedure consists in sampling definite grouping of leaves, but with random selection of stalks. Two sampling stations are established for each field. Two disks are removed from each sampled leaf and sixty leaf disks constitute a sample. One sample is taken from each station. Repeated checks with this procedure have indicated that where growth in a field is uniform, the results obtained for the two stations are nearly always in very close analytical agreement. This apparent uniformity of nitrogen and plant relationship within a field, the simple system of leaf sampling, and precision of analysis have established a foundation which, it is hoped, may be found suitable for purposes of establishing a satisfactory field control.

A pot experiment was conducted wherein a relationship was found to exist between the nitrogen content of the leaf samples and elongation of the stalk. The method of determining nitrogen index in the cane plant, as described in this paper, was applied to a number of field surveys from which it was determined that the nitrogen content of the field samples were within the same range as those determined in potted cane. A study of growth data of field and potted canes indicated that there is also a similarity in the relationship between fluctuation of nitrogen content in the leaf system at periodic intervals and growth of sugar cane in the corresponding intervals.

A study of the data indicates that apparently a critical nitrogen percentage in the leaf-punch sample may be established, above which a state of luxury consumption is indicated. The optimum nitrogen index lies between the critical high point and a minimum value, below which growth as measured by the elongation of stalk is negligible. When the nitrogen data, termed as "nitrogen index" values, are plotted with the indices on the Y-axis and the monthly intervals of collection on the X-axis, the resulting curve will provide a graph showing the progress of the nitrogen situation in the entire crop. Under normal conditions, this curve can be influenced readily by fertilization and advantageously so at several intervals of growth.

From results of this study, levels or limits of the nitrogen content of the leaf-punch samples have been tentatively prescribed for a 24-month crop of cane. It is proposed to interpret the leaf nitrogen data with respect to the sufficiency of the

nutrient for the crop throughout its growth cycle. These limits and interpretations are incorporated into a tentative method for the control of nitrogen fertilization. In brief the control may be achieved by collecting leaf-punch samples at progressive intervals, obtaining their total nitrogen content (nitrogen indices) by analysis with a rapid chemical method and determining the need of the crop for nitrogen from a study of the plotted data. Practical applications are discussed and illustrated with data presented graphically.

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## Dead Cane at Harvest

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By J. P. MARTIN

The subject of dead cane at harvest has been investigated on different occasions, but more recently it has been brought up by H. P. Agee for further consideration. The object of this paper is to present and briefly discuss those factors responsible for dead cane at harvest, a number of which have been suggested by persons interested in the general subject. It is hoped that this article will stimulate a wider interest in this important phase of sugar cane culture and that contributions from others will be received.

Little difficulty is experienced in maintaining an accurate record of all existing conditions in a cane field from the time sugar cane is planted until it begins to lodge. However, once the cane has become lodged, especially with the heavy tonnages which result in a thick blanket of cane and trash, it becomes most difficult to record accurately the conditions within a field, many of which are harmful and become apparent only at harvest.

The age of cane at harvest is usually between 14 and 24 months, although in some instances the age may be less than 14 months or more than 24 months. The environment, variety, field culture, economic conditions, pests, and diseases are some of the major factors which govern the age of cane at harvest and which are responsible for a certain amount of dead cane. It is not uncommon to observe dead cane at harvest; at times the amount present causes a marked reduction in yields, again in some fields little or no dead cane occurs. Dead cane is more apparent where the crop is hand cut than where it is mechanically harvested.

If one is cognizant of existing environmental conditions during the growth of a field and aware of the factors which cause cane to die prior to harvest he is in a much better position to render a satisfactory explanation of the problem. With such a knowledge it may be possible to adopt and carry out definite measures for maintaining conditions favorable to the development of the cane plant and thus reduce monetary losses.

It might be desirable to consider the individual stalk as the unit in a field rather than the stool since field observations at harvest have shown that it is the aggregate of individual stalks that makes up the major portion of dead cane. Under some circumstances two or more dead stalks may occur in a stool but seldom does the entire stool die. A careful examination of those dead stalks which are found at harvest has, in the majority of cases, shown that the stalks made a normal growth for a certain period as evidenced by the length and diameter of internodes and total stalk length. Often these growth indices, when studied in relation to mature cane, aid in establishing approximately the time the stalks died. For example, short, medium, or long, dead stalks would indicate that such stalks had died during the early, middle, or latter part of the crop, respectively.

*Environment:*

The environment includes "all the external conditions and influences affecting the life and development of an organism." We may think of the environment of the cane plant as being made up of the climatic and soil factors. The climate is determined by such factors as light, temperature, rainfall, humidity, and wind. In considering the soil we must think of the physical factors, namely, texture, structure, tilth, depth, aeration, moisture retention, drainage, etc., and the chemical factors such as soil acidity, and the chemical composition of the soil and of the soil solution. All of the above factors govern to a large extent the physiological processes taking place within the plant; any one factor if unfavorable to the plant may retard its growth and cause it to die before reaching maturity.

In order to appreciate the effect of climate on cane growth one has only to go from the lower to the higher elevations on any plantation. Possibly the two climatic factors most responsible for the heavier yields at the lower elevations are increased sunlight and higher temperatures.

We realize that the total number of shoots which start in a plant or a ratoon field never reach maturity. R. J. Borden called the writer's attention to some very interesting data on stalk mortality in unpublished studies which he and F. C. Denison conducted in 1930 and 1931 with H 109 and POJ 2878 at Waipio Substation. The average per cent mortality of cane stalks at different periods from seven plots (each plot .0861 acre in size) of each variety and treatment, as determined by stalk counts follows:

Variety	Between 2nd and 5th months	Between 5th and 9th months	In first 9 months	Between 9 months and harvest	Stalks counted at 2 months which failed to survive until harvest
H 109*	13.1±7.8	10.1±3.0	22.1±6.5	22.6±3.8	39.5±5.8
POJ 2878*	21.9±9.7	8.5±2.2	28.7±8.4	7.1±3.3	31.9±8.5
H 109†	30.5±4.5	14.9±3.1	41.1±1.8	25.4±6.1	56.7±3.8
POJ 2878†	37.5±8.1	16.6±2.0	48.1±5.9	17.3±4.0	56.7±6.5

\* Cane planted in standard furrows, 5 feet apart.

† Cane planted in borders 10 feet wide, rows 3.3 feet apart.

In more recent studies Mr. Borden offers the following figures regarding stalk mortality in 31-1389, plant cane, Field 19, Makiki Station:

SHOOTS PER FOOT OF CANE LINE  
(Planted April 13, 1937)

July 30 10.6	August 30 5.9	May 1, 1938 at harvest 4.6
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From the above figures it is very apparent that a high mortality of cane stalks occurs shortly after the cane begins to close in, partly as a result of reduced light—the shoots having been "shaded out." A certain amount of this mortality is probably due to competition and crowding of stalks within the stool. The death of the young shoots at this period of growth is not greatly reflected at harvest merely because an acre of land can only support or accommodate, depending on the variety, in the neighborhood of some 30,000 or 40,000 mature stalks. During the growth of a crop dead stalks are observed where cane has become lodged by wind, by the

practice of pushing back, or as the result of its own weight, and the only apparent cause of death is reduced light or a shading out of the growing top; dead cane from this cause cannot be diagnosed at harvest.

It is not uncommon to come upon depressed cane growth or even dead cane in regions exposed to long periods of severe drought or in localized areas of excessive soil moisture. These adverse conditions may occur at any time before the crop reaches maturity. In regions of heavy rainfall, conditions are favorable for the development of various stalk rots which result in a certain amount of stalk deterioration; under dry conditions these rots are less active.

Light showers and high relative humidities are not responsible for dead cane but they do favor the rapid spread and development of eye spot disease which when severe often produces top rot; if the stalks manifesting top rot fail to send out side shoots or "lalas" they soon die. In fields so affected or where individual stalks scattered throughout the field have been killed, then an appreciable amount of dead cane at harvest can be expected.

During severe wind storms large areas of cane become flattened as though a large roller had passed over the fields; this condition is more pronounced when strong winds are accompanied or preceded by heavy rains. Cane stalks injured by wind frequently break near their base and may continue to live for several months. Strong winds may cause a sudden drying of the leaves or a breaking of the cane top near the growing-point region and in either case the stalk sometimes dies. During one wind storm the cane may be lodged in one direction and during the next storm, if the wind is from an opposite direction, the cane may be lifted and laid in a different direction; a sudden shifting of cane at any period of its development, but especially toward maturity, causes dead cane which at harvest cannot be easily explained. When wind damage is observed it might be advisable to mark such areas and attempt to measure the amount of dead cane at harvest. The brittleness of a variety has a direct bearing on the number of broken and dead stalks following a wind storm. A record of field conditions where portions of a field have suffered from wind damage might explain why more dead cane at harvest occurs only in certain parts of the field.

The physical and chemical factors of a soil may at times play a part in the problem of dead cane at harvest. We refer particularly to acute cases of excesses of soluble salts of sodium and magnesium in the soils, or deficiencies of one or more of the essential elements. Cane plants suffering from malnutrition usually manifest leaf or stalk symptoms by which such nutritional disturbances may be recognized. Possibly these factors, if unfavorable for cane, play a greater part in relation to depressed or abnormal growth than they do to dead cane at harvest.

#### *Varieties:*

Under a given set of environmental conditions the ripening period of one variety sometimes varies from that of another and the holding-over qualities may also vary. Yellow Caledonia, for example, after reaching maturity is able to retain its juice quality for a much longer period than D 1135. Early maturing varieties with poor holding-over qualities deteriorate rapidly and may develop a condition known as sour



rot, the final result of which causes the entire stalk to die. These factors are of considerable importance in relation to dead cane at harvest only when cane is held for some time after it reaches maturity.

The qualities of a variety, namely, hardness of rind, tasseling, size of stalk, rate of growth, erect habit, and secondary growth are of primary importance in relation to those factors producing injuries to the cane plant. Where the cane borer is a problem it is highly desirable to have a variety with a hard rind. Varieties that tassel freely but do not readily develop lalas are undesirable since tasseled stalks without lalas die. The size of stalk, rate of growth, and erect habit of a variety have a direct bearing as to the time cane begins to lodge. Heavy secondary growth, as produced by some varieties, covers and shades out a number of primary stalks which have become lodged with the result that the latter die prior to harvest. A splitting of internodes known as "growth cracks" develops in a few varieties; this condition is undesirable since it permits the entrance of organisms which may cause the stalks to sour or rot.

### *Field Culture:*

A greater efficiency in supervision and irrigation has been obtained by the practice of "pushing back" cane along irrigation ditches. If the cane is pushed back at a time when the soil is wet and soft from rains or irrigations little or no damage to the plant results; however, if this practice is not carefully carried out considerable stalk and root injury takes place. In order to reduce stalk damage to a minimum the cane should be pushed back gradually, otherwise an abnormal amount of dead cane may be present along ditches when the field is harvested.

A small degree of damage to the cane plant occurs during various field operations, *viz.*, disking, cultivating, weeding, etc. In the early part of the crop these injuries are of little significance but they may permit the entrance of parasitic organisms which frequently cause a souring or rotting of the stalks. The tractors used for drawing mechanical implements in a field break and injure a considerable number of stalks.

Some injury to well-formed and mature stalks has been caused by chemical weed sprays. This is especially true along edges of fields where weed sprays are applied repeatedly and the spray comes in contact with the stalk. Usually the damage extends only a short distance within the stalk but in some instances practically all the stalk tissue at the base is killed and the stalk eventually dies.

In a harvested field where "high cutting" is in evidence a certain amount of dead cane can be expected when the field is again harvested. The primary shoots in a ratoon crop develop from the underground buds of the previous crop. The buds on exposed stubble or high-cut cane develop at first into what may be considered normal shoots. Later if such stalks are examined the point of attachment to the stubble will be found to be extremely weak and one can realize how easy it is for the stalk to break at this point. Furthermore it is difficult to "hill up" high-cut cane sufficiently to overcome this weakness. A careful study of individual dead stalks at harvest showed definitely that a number had developed from stubble of high-cut cane of the previous crop. These observations were made by following individual

dead stalks to the ground level where they were found to be broken but still attached to the exposed stubble. Similarly weak stands may come about (especially with varieties that lack vigorous ratooning properties) when overcast weather prevails at the start of a crop, or when weed growth shades the soil, so that the sun does not warm it to sufficient depth to bring about the sprouting of deeply placed eyes of the stubble. In consequence, there is reason to believe that the stand of cane is made up largely of shoots which developed from near the surface. Many of these surface shoots break off and die when the crop lodges.

Observations to date indicate that there is less dead cane in plant than in ratoon crops. In plant fields there is sufficient soil around the base of the plants for considerable mechanical support and the stalks are firmly attached to the underground portion of the stool, whereas in ratoon fields, especially with high-cut cane, the opposite conditions are frequently found.

A short-cropping system tends to make for less dead cane at harvest than a long-cropping practice simply because the cane after it has become lodged is exposed for a much shorter period to those detrimental factors, causing cane to die prior to harvest.

In some instances an insufficient amount of the necessary fertilizer may result in depressed cane yields but rarely does cane die from this cause. On the contrary, excessive fertilization with nitrogen tends to produce soft rank growth that is more susceptible to the hazards that cause dead cane. In a few specific localities where an acute deficiency of an element exists the cane may die before it reaches maturity unless the necessary element is made available for the plant.

The aim of all field cultural practices is to have the maximum number of mature stalks in a sound condition at harvest.

#### *Economic Conditions:*

The age of cane at harvest depends on a number of conditions, such as, climate, the variety, soil, irrigation, fluming water, fertilization, time of planting, cultural practices, pests, diseases, etc. In recent years certain economic conditions have made it impossible to harvest some fields at the optimum age with the result that an excessive amount of dead cane was present at harvest.

#### *Insects:*

C. E. Pemberton points out that the beetle borer, *Rhabdocnemis obscura* (Boisd.), prefers recumbent to standing cane and that its damage is more severe in long crops than in short crops. The runways or tunnels made by the grub of the beetle borer are often very extensive and they not only weaken the vitality of the stalk but may cause it to break. Red rot disease and a souring of the stalk by fermentative organisms often follow borer injury and contribute greatly to economic losses. The beetle borer is probably one of the factors causing cane to die before it reaches maturity. It is definitely known however to be attracted to and develop in dying or dead cane and its exact importance as a causative factor in the formation of dead stalks is not fully determined.

The grub of the Anomala beetle, *Anomala orientalis* (Waterh.), feeds on the underground portion of the cane plant and through its injuries parasitic fungi and bacteria sometimes invade the plant and produce a rotting or souring of the stalk. According to Mr. Pemberton a certain number of dead stalks may in some regions be attributed to Anomala injury followed by parasitic organisms; when 100 or more grubs appear in single stools, root pruning may be so severe that most of the stalks in the stool die.

In some instances soil-inhabiting animals, for example two species of nematodes, *Tylenchus similis* Cobb, and *Heterodera marioni* (Cornu), have produced considerable injury to cane roots and have been responsible for reduced cane growth and possibly have contributed toward an early death of cane stalks.

#### *Diseases:*

The leaf, leaf and stalk, stalk, and root diseases which sometimes cause cane to die prior to harvest are discussed separately.

Very few diseases attacking only the leaves actually cause the death of the plant. Of these diseases limestone chlorosis and Pahala blight when severe have been responsible in a few instances for the death of a small amount of cane.

The greatest amount of dead cane occurs from the leaf and stalk diseases which are given in their order of relative importance: eye spot, leaf scald, red rot, pokkah boeng, and red stripe.

The relation of eye spot to dead cane at harvest has been mentioned under *Field Culture*. It might be stated that eye spot in the last two decades has caused more cane to die before harvest than any other disease. Eye spot when present is most severe in low-lying areas where the cane is making a rapid growth and where conditions are as a rule more favorable for the other harmful factors.

The severity of leaf scald is governed to a large extent by existing environmental conditions and during epidemics of the disease cane is often killed outright. The greatest losses usually take place when cane growth is retarded as a result of low soil fertility, drought, or maturity. Cane killed by leaf scald can often be recognized at harvest by the presence of the numerous side shoots or lalas on the dead stalks.

Red rot brings about the death of many stalks; the red rot organism enters the stalk through animal, insect, chemical and mechanical injuries and at times losses are of major importance.

An occasional cane stalk has been killed by pokkah boeng but as a rule affected plants recover. In localities where red stripe is serious, top rot results and unless the lateral buds develop the affected stalks die from the effects of the disease. Red stripe is primarily a leaf disease but in some instances it affects the spindle, growing point and stalk. Pineapple disease has caused some losses in standing cane but such cases are rare.

The root diseases responsible for dead cane are *Pythium* and *Marasmius* root rot. Cane in a weakened condition may die from *Marasmius* root rot; the disease itself is considered of secondary importance inasmuch as it attacks cane that is in a poor growing condition. With the commercial varieties grown today *Pythium*

root rot is important only where malnutrition is a contributing factor; however, with varieties susceptible to *Pythium* root rot an excessive amount of dead cane at harvest can be anticipated.

### *Various Injuries:*

Under this general heading some of the more important miscellaneous injuries to sugar cane should be mentioned.

We all fully appreciate the seriousness of the rat problem and the large economic losses resulting therefrom. The injured stalks are not only weakened in vigor but they often break at the point of injury and die. The large wounds caused by rats permit the entrance of pathogenic organisms which cause a rapid souring and rotting of the stalks. Rat injury is commonly found on dead cane when examined at harvest and is no doubt one of the largest contributing factors to this general problem.

Although of minor significance fire burn, leaf burn, and lightning injury are three elemental injuries which have caused cane to die in a few instances. Cane fields located close to the ocean are sometimes injured by salt spray and a small amount of cane has died from frequent exposures to salt spray.

Chemical injuries from fertilizers and weed sprays play a small part in the general subject; the latter has been discussed under *Field Culture*. Fertilizer burn is of more concern with young cane since practically all of the fertilizers are applied during the first half of the crop cycle.

In discussing the subject of dead cane at harvest Mr. Agee has submitted the following comments:

It may be said that while the importance of none of the specifically known causes are to be discounted in accounting for dead cane at harvest (such as wind, rats, borers, diseases, etc.) yet we must concede that apart from these there are death factors of high importance that are not sufficiently well understood. Self-shading or lack of sunlight due to lodging or to heavily overcast weather for a prolonged period no doubt play a prominent part in killing many stalks or in weakening them to a point where they easily succumb under the influence of other factors that contribute to their death.

A certain amount of dead cane at harvest is likely to come about inevitably with high cane tonnages. The life span of the individual stalk of cane will vary enormously under differences in environment and in accordance with varietal influences. Scores of cane varieties fail to find acceptance because stalk mortality is too high; the few varieties that do become commercial ones succeed because of a vitality that keeps so large a part of the stalks alive to the point of harvest. A variety that succeeds in one environment fails often in another largely because so many stalks die in advance of harvest. H 109, for instance, is a cane variety that thrives under conditions of high sunlight; stalk mortality is high if it is grown in areas of high rainfall and overcast skies. This variety, grown in areas that are only fairly well suited to it, is apt to show a high sensitiveness to climatic variations. In bright years it does well, in dull years it may do poorly.

As Dr. Mangelsdorf has pointed out, the conditions that permit of our high cane yields under long crops, 20 to 24 months, are rather special ones. In few places other than Hawaii do these conditions prevail.

In the warmer environment of Java, cane on lodging loses its sugar content rapidly and deteriorates badly. The stalks send out roots and lalas, many of them die.

The low lands of Oahu and Maui have the conditions of moderate heat and bright sunlight that are well suited to heavy cane yields under long crops, and these when we come to analyze it are conditions suitable to cane stalk longevity.

In other places, certain fields of windward Kauai and Kohala are examples, cane makes good growth the first twelve months, lodges and thereafter, probably through lack of the temperatures of the better irrigated lands, fails to make growth the second year in keeping with that of the first. It would seem, though we lack the specific knowledge we would like to have to support the theory, that when a field of cane lodges and the cane tops lose the positions they have held in the sunlight, a considerable amount of vegetative vigor is needed for the stalks to gain new positions that give them the light they need. Cane stalks reach for light and move into it by the wedge-like growth that takes place in meristematic tissue of each joint. By this means they bend in one way or another to find a place for their leaves in the sun.

Under a lack of temperature (or light) that induces the required vigor to bring about this adjustment of position effectively, self-shading, under the stress of lodging, probably becomes a potent factor in stalk mortality.

Consider that a plausible explanation rather than a proved fact, and it leads us to the point of saying that stalk mortality at harvest will be better understood when we have a better understanding of the whole gamut of environmental influences as they affect the crop as it grows to a heavy stand, as it thins down the number of young shoots by self-shading, as it inclines or lodges and readjusts itself as best it can to these trying circumstances, and as it continues growth to harvest.

The precise study of how the cane plant, as part of a cane crop, lives and grows, and thrives or dies, is a subject of inviting interest, and is furthermore one in which all of those who tend cane fields can participate, to the advantage, quite likely, of themselves and the industry as well.

Succinctly expressed we often know just why cane stalks die, but there are many occasions of high mortality where the death factors are surrounded with a great deal more mystery or uncertainty than we can be content to tolerate.

#### *Acknowledgment:*

The writer is indebted to Messrs. H. P. Agee, R. J. Borden, C. W. Carpenter, A. J. Mangelsdorf and C. E. Pemberton of this Station for their valuable contributions which have been included in this paper.

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## The Effects of Oven Drying and Air Drying on the Available Nitrogen Content of Soils

By P. E. CHU AND FRANCIS E. HANCE

At the time a field crop of sugar cane is harvested the soil in the field, as a rule, is quite low in its concentration of "available" or readily soluble nitrogen. Immediately thereafter, however, upon exposure of the bare field to sunlight (warmth) and moisture, bacterial action appears to be stimulated and the formation of available nitrogen occurs from insoluble organic sources in the soil. Fukunaga and Dean (2) describe this process as "mineralization of nitrogen."

As a companion determination to the progressive sugar cane leaf-punch nitrogen field survey, an appraisal of the status of available soil nitrogen, at any given moment, is highly desirable. It is common knowledge, however, that by the time representative soil collections can be made, dried and composited for analysis a delay of ten days or longer will have ensued and the available nitrogen concentration will have been markedly changed. Either one of these conditions defeats the purpose of the determination.

Therefore an attempt has been made (a) to ascertain the shift in soil nitrogen availability as brought about by various methods of artificial and natural drying of the soil specimen, and (b) to develop a rapid method of measuring soil nitrogen availability with fair accuracy in the shortest possible space of time immediately following the sampling of the field soil.

As a general rule, soils intended for analysis are air dried before they are disintegrated, sieved, mixed and prepared for the analyst. Soils taken from the field may vary from saturation with moisture to a wetness below the wilting coefficient. On some plantations, or in localities of high humidity, air drying of some soils may require days or even weeks. Hence, if drying is an analytical prerequisite, rapidity of drying is a necessity unless other methods of making the determination be found.

Obvious methods of drying soil are to place the sample in an oven at controlled temperature or near sources of warmth in the sugar factory. We shall consider the extent of the changes, if any, that take place in the ammoniacal, nitrate and *total available* nitrogen content of soils when they are dried by various means and for different periods of time.

Russell and Petherbridge (7) found that the rate of decomposition of organic matter by bacteria and the formation of ammonia increases, with increased temperature, under moist conditions and reaches a maximum of about 45° C. Lyon and Bizzell (5) found that water-soluble nitrogen increases when soils are sterilized or are placed in the oven at 100° C.

Alexander (1), Lyon and Bizzell (5) and Webster (8) also reported that plants grow better in steamed soils after a certain period. Russell and Hutchinson (6) found that partial sterilization causes a fall in the number of soil bacteria and soon after an increase takes place, together with a rise in the ammoniacal fraction. Fukunaga and Dean (2) report that they note an initial rapid release of nitrogen apparently associated with microbiological decomposition of nitrogenous compounds when soils are incubated. This is followed by a second phase of slow release.

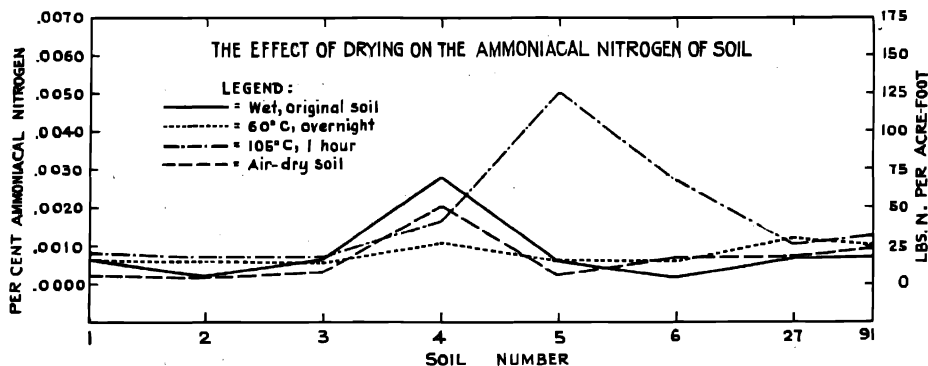


Fig. 1

### EXPERIMENTAL

Temperatures were determined at which it was found possible to dry soils in a short time, say, between an hour and twenty-four hours. Analyses of the samples were made immediately after drying by means of the rapid chemical methods (3, 4). This procedure would indicate any increases of available nitrogen which may have developed due to heating and also it should show the extent of such changes as brought about by the heating.

Preliminary tests with soils Nos. 27 and 91 indicate that at 105° C. these samples were sufficiently dry to be workable in about one hour. At 60° C., twenty-four hours or longer are necessary to secure comparable drying. These two temperatures were therefore selected to produce what may be considered good indications of the changes which may take place in the available nitrogen of soils when so treated. For this purpose a representative number of soils were selected which are known to be difficult to dry.

In order to study the effect of prolonged heating at these temperatures, other portions were dried at intervals in the oven, some for as long as two continuous weeks. Comparative figures were also obtained on air-dried and wet portions of these soils. The air drying was carried out in the shade at room temperature. The method used to sample the wet soil is described in the appended procedure.

The analytical data are discussed separately under (a) ammoniacal, (b) nitrate, and (c) the sum of the two, or total available nitrogen.

#### *Ammoniacal Nitrogen:*

Comparisons of the ammoniacal nitrogen content of the soils studied show that the air-dried portions are, in most cases, slightly lower in ammonia nitrogen than the moist, original specimens (Fig. 1 and Table I). The changes found were less than 25 pounds per acre-foot in the eight soils studied. Minor differences were also noted when the soils were heated overnight in an oven below 60° C. These soils were not completely or sufficiently dried. One sample, No. 4, decreased by 40 pounds ammonia nitrogen per acre-foot compared to the wet soil, but this ammonia was apparently nitrified and not lost, for the nitrate nitrogen increased by the same amount. When the drying period was advanced to 40 hours, there were small gains observed generally. However, samples kept in the oven at 60° C. for two weeks increased tremendously in ammonia nitrogen in every case. The gains ranged from 65 pounds to 300 pounds nitrogen per acre-foot.

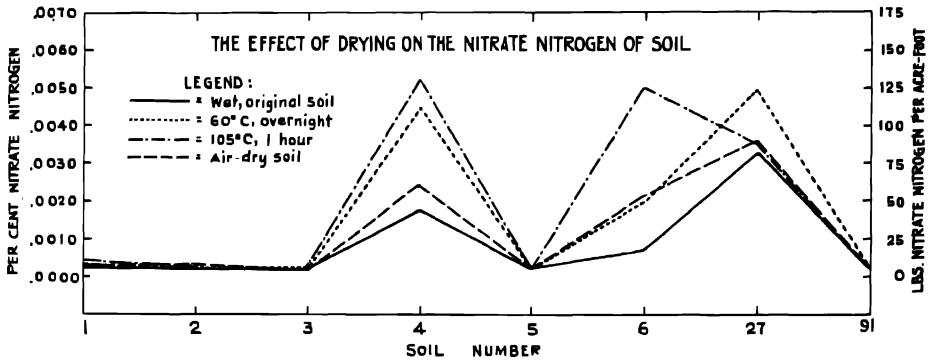


Fig. 2

On drying for one hour at 105° C., the changes in the ammoniacal nitrogen content varied from a 30-pound decrease to an increase of over 100 pounds (Soil No. 5). The decrease in the one soil (No. 4) is traceable to nitrification which also occurred at 60° C. However, when the soils were left in an oven over a weekend (about 65 hours) at the higher temperature, significant increases of 50 pounds to over 200 pounds nitrogen per acre-foot took place in every case. These figures show that an increase in temperature and also in the period of heating markedly step up the concentration of ammoniacal nitrogen in these soils. Below 60° C., where it is necessary to dry for twenty-four hours or longer, slight to over 25-pound increases of ammonia nitrogen were found. At the higher temperature of 105° C. for one hour, very great changes may thus be expected in some soils. For this reason temperatures as high as 105° C. can not be used to dry soils for the determination of field availability in ammoniacal nitrogen. Extended heating at elevated temperatures is especially to be avoided, even temperatures as low as 50° C. to 60° C. Under these conditions the factors favoring ammonification are intensified and predominate and large increases of ammonia generally prevail. Such conditions, of course, do not exist in the fields.

#### *Nitrate Nitrogen:*

The nitrate form of nitrogen is generally not affected much by temperature changes or by extended heating. In the two soils, Nos. 4 and 6 (Table II and Fig. 2), in which large increases of nitrate nitrogen do occur upon heating, it may be explained, perhaps, as due to the acceleration of the natural processes of nitrification. By subsequent re-analyses of all samples, the wet soils were found to have increased in nitrate nitrogen to the high levels reached by the samples dried by the various methods described. In soil No. 4, the gain is apparently due to nitrification of the ammoniacal nitrogen originally present in the wet soil and in No. 6, to nitrification of organic matter.

Nitrification appears to be accelerated in the early stages of heating and ammonification in the later stages.

#### *Total Available Nitrogen:*

Total available nitrogen, i.e., the sum of the ammoniacal and the nitrate forms, in naturally wet soils appeared to change only slightly, if any. Exceptions were



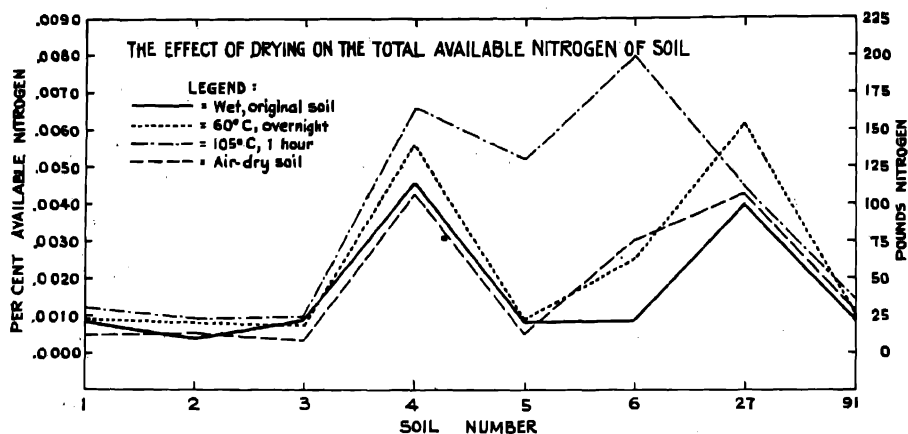


Fig. 3

found, however. In one wet soil, No. 6 (Table III and Fig. 3), the total available nitrogen increased by about 50 pounds per acre-foot when the soil was air dried. This gain is due to an increase of both the ammoniacal and nitrate forms. After standing for two weeks, this particular soil gained as much as 40 pounds nitrate nitrogen. It is further observed that this soil kept in a moist condition for a period of time increased to the same nitrogen content as the air-dried portion when both were re-analyzed later. The conclusion which may be drawn safely, we believe, is that the analysis of a soil for available nitrogen in naturally wet specimens taken from the field is quite possible and gives reasonably true and accurate results. Where an apparent exception is found, as was the case in soil No. 6, it may be due to an actual change taking place during the time required to air dry the soil.

When dried overnight in an oven at 60° C., or lower, gains up to 50 pounds nitrogen per acre-foot were found. When the soils were left over a weekend in the oven at this temperature, increases ranged up to 100 pounds nitrogen per acre-foot. Samples kept for two weeks at 60° C. gained from 65 pounds available nitrogen to over 300 pounds. At a temperature of 105° C. for one hour, these soils gained from 3 pounds to over 175 pounds nitrogen and increased from 55 pounds to over 225 pounds when incubated (dried) over a weekend at 105° C.

Drying in the oven either at 60° C. or at 105° C. increases the available nitrogen content of these soils. The increases which take place in a sizable proportion of the samples dried at 60° C. and for a short period occur in every soil tested when dried for longer periods or at higher temperatures.

#### DISCUSSION

The experimental figures, although obtained from a limited number of soils which were nevertheless a representative selection, definitely support the belief that analyses of wet field samples of soils give a truer picture of the available nitrogen supply in the field at the time of sampling than either air-dried or oven-dried portions of the same soils. This brief study was not intended to learn the causes governing the relationships found to exist between nitrogen values and various methods of soil drying, but there are sufficient data presented to point toward accelerated bacterial activity and also chemical decomposition due to the

elevated temperatures employed as major causes of the increases in available nitrogen noted in the dried portions of the soils. Soils which were dried, either oven or air dried, when left standing in the dry state for a week to ten days did not change in available nitrogen as compared to their respective contents immediately after drying. (Refer to the lower figures in Tables I, II and III.) However, when the dried soils were kept moistened with distilled water for a week, the portions dried at 105° C. and also those dried at 60° C. for periods longer than 40 hours showed tremendous increases of available nitrogen. Although no attempt was made to keep the soils uncontaminated, those kept at 105° C. for 60 hours are apparently only partially sterilized. On the addition of moisture, the bacteria which produce ammonia increased to the greatest extent in the soils kept at the highest temperature and for the longer period. This is shown (Table I, upper figures) in the ammonia content of the soils. The organisms detrimental to the nitrifying bacteria were not affected or destroyed when heated below 60° C. overnight.

The soils selected for this test included those extremely difficult to dry. The wet sampling method proposed is admittedly difficult to employ on these soils, but when these same soils were dried, by whichever means selected, the subsequent handling of the dry specimens was much more difficult and consumed more time than the method suggested. This is due to the fact that the soils in question caked into a solid, rocky mass when dried.

The laboratory sampling method used, and which is recommended for handling moist or wet field soils, is as follows:

1. If the sample taken from the field is large, spread the soil out on a wide sheet of heavy paper.
2. Break up the large masses into smaller pieces with a trowel.
3. Start from one end of the sheet and quarter by spading off a quarter of each large mass of soil and also taking one-fourth of the smaller pieces.
4. Break up the sample further and repeat Step 3 until one to two pounds are obtained.
5. Spread the sample on a square, 8-mesh wire screen (8 mesh to a linear inch) about 18 inches to a side. Press the soil with a large wooden mallet. (The screen should be made so that the wire bottom rests about three inches above the table, the screen height being about two inches or more.)
6. Press a portion of *each* part of the sample through the screen until about a tumblerful is obtained. (It is not necessary to press all of the soil through the screen.)
7. Mix by means of a spatula.
8. Fill the 10-gram soil cup by taking a small portion of soil from various parts of the mound, pack the cup solidly and level it.
9. Remove excess soil from spatula and cup.
10. Transfer the contents of the cup to a 250-ml. beaker by digging out neatly with the cleaned spatula.
11. Add 50 ml. Reagent 5 N and stir well, using two stirring rods held together in one hand if necessary.
12. Filter through a dry filter into a 100-ml. beaker and proceed with the usual R.C.M. ammoniacal and nitrate nitrogen determinations on the filtrate.

TABLE I  
PER CENT AMMONIACAL NITROGEN

Lab. No.	Description of samples	Specimens analyzed	Treatments									
			Air-dry soil	Wet orig. fld. soil	Over-night	Below 60° C.				105° C.		
						40 hrs.	2 wks.	1 hr.	60 hrs.	1 hr.	60 hrs.	
1	Honolulu Plantation Co., near poi factory	{ Immediately after treatment After 2 weeks	.0002 .0002	.0006 .0002	.0006 .0002	.0007 .0006	.0050 .....	.0008 .0006	.0028 .....	.0008 .0006	.0028 .....	
2	Expt. Stn. Seedling Station, Ewa	{ Immediately after treatment After 2 weeks	.0002 .0002	.0002 .0002	.0006 .0003	.0009 .0007	.0028 .....	.0007 .0006	.0026 .....	.0007 .0006	.0026 .....	
3	Waialua Agric. Co., Ltd., Mokuia side	{ Immediately after treatment After 2 weeks	.0002 .0004	.0007 .0002	.0006 .0006	.0008 .0006	.0056 .....	.0007 .0007	.0028 .....	.0007 .0007	.0028 .....	
4	Waialua Agric. Co., Ltd., valley soil	{ Immediately after treatment After 2 weeks	.0019 .0021	.0028 .0002	.0011 .0011	.0020 .0018	.0150 .....	.0016 .0018	.0100 .....	.0016 .0018	.0100 .....	
5	Kahuku Plantation Co., Waialua side	{ Immediately after treatment After 2 weeks	.0003 .0002	.0006 .0002	.0006 .0003	.0007 .0006	.0005 .....	.0005 .0050	.0030 .....	.0005 .0050	.0030 .....	
6	Kahuku Plantation Co., Punaluu	{ Immediately after treatment After 2 weeks	.0008 .0006	.0002 .0002	.0006 .0003	.0012 .0010	.0100 .....	.0030 .0024	.0050 .....	.0030 .0024	.0050 .....	
27	Olaa Sugar Co., Ltd. Field 4-5	{ Immediately after treatment After 2 weeks	.0007 .0012	.0007 .....	.0012 .0011	.0000 .....	.0000 .....	.0010 .0010	.0050 .....	.0010 .0010	.0050 .....	
91	Manoa Substation, surface, Manka of D-3	{ Immediately after treatment After 2 weeks	.0009 .....	.0007 .....	.0010 .0008	.0000 .....	.0000 .....	.0012 .0011	.0100 .....	.0012 .0011	.0100 .....	

\* Dried on hot plate for 10 hours.

Plus sign (+) denotes quantity more than that indicated.

TABLE II  
PER CENT NITRATE NITROGEN

Lab. No.	Description of samples	Specimens analyzed	Treatments					
			Air-dry soil	Wet orig. fld. soil	Over-night	Below 60° C.	105° C.	
1	Honolulu Plantation Co., near poi factory	(Immediately after treatment) (After 2 weeks)	.0003 .....	.0002 .0005	.0003 .0002	.0005 .0002	.0004 .0002	.0002 .0002
2	Expt. Stn. Seedling Station, Ewa	(Immediately after treatment) (After 2 weeks)	.0003 .....	.0002 .0005	.0002 .0002	.0002 .0002	.0002 .0002	.0002 .0002
3	Waialua Agric. Co., Ltd., Mokuleia side	(Immediately after treatment) (After 2 weeks)	.0002 .....	.0002 .0003	.0002 .0002	.0002 .0002	.0003 .0002	.0002 .0002
4	Waialua Agric. Co., Ltd., valley soil	(Immediately after treatment) (After 2 weeks)	.0024 .....	.0018 .0050	.0045 .0040	.0040 .0045	+.0050 .0040	.0028 .0028
5	Kahuku Plantation Co., Waialua side	(Immediately after treatment) (After 2 weeks)	.0002 .....	.0002 .0002	.0003 .0002	.0003 .0002	.0003 .0003	.0002 .0002
6	Kahuku Plantation Co., Punaluu	(Immediately after treatment) (After 2 weeks)	.0022 .....	.0007 .0022	.0017 .0020	.0035 .0031	+.0050 .....	.0030 .0030
27	Olaa Sugar Co., Ltd. Field 4-5	(Immediately after treatment) (After 2 weeks)	.0036 .....	.0033 .....	.0050 .0045	.....	.0035 .0033	.0038 .0025*
91	Manoa Substation, surface, Mauka of D-3	(Immediately after treatment) (After 2 weeks)	.0002 .....	.0002 .....	.0002 .0002	.....	.0002 .0002	..... .0002

\* Dried on hot plate for 10 hours.

Plus sign (+) denotes quantity more than that indicated.

TABLE III  
PER CENT TOTAL AVAILABLE NITROGEN

Lab. No.	Description of samples	Specimens analyzed	Treatments					
			Air-dry soil	Wet orig. fld. soil	Over-night	Below 60° C.		
						40 hrs.	2 wks.	105° C.
								1 hr. 60 hrs.
1	Honolulu Plantation Co., near poi factory	Immediately after treatment	.0005	.0008	.0009	.0012	.0053	.0012 .0030
		After 2 weeks	.0005	.0007	.0004	.0008	.....	.0008 .0026
		Water added after heating	.....	.0008	.....	.0027	.....	.0042 .0072
2	Expt. Stn. Seedling Station, Ewa	Immediately after treatment	.0005	.0004	.0008	.0011	.0030	.0009 .0028
		After 2 weeks	.0004	.0007	.0005	.0009	.....	.0008 .0023
		Water added after heating	.....	.0010	.....	.0028	.....	.0042 .0102
3	Waialua Agric. Co., Ltd., Mokuleia side	Immediately after treatment	.0004	.0009	.0008	.0010	.0059	.0010 .0030
		After 2 weeks	.0006	.0005	.0008	.0008	.....	.0009 .0024
		Water added after heating	.....	.0022	.....	.0042	.....	.0062 .0102
4	Waialua Agric. Co., Ltd., valley soil	Immediately after treatment	.0043	.0046	.0056	.0060	+.0170	+.0066 +.0130
		After 2 weeks	.0044	.0052	.0051	.0063	.....	.0058 +.0130
		Water added after heating	.....	.0052	.....	.0105	.....	.0105 .0122
5	Kahuku Plantation Co., Waialua side	Immediately after treatment	.0005	.0008	.0009	.0010	.0053	+.0053 .0032
		After 2 weeks	.0004	.0004	.0005	.0008	.....	.0053 .0028
		Water added after heating	.....	.0015	.....	.0040	.....	.0108 .0082
6	Kahuku Plantation Co., Punaluu	Immediately after treatment	.0030	.0009	.0025	.0047	+.0130	+.0080 +.0080
		After 2 weeks	.0024	.0024	.0023	.0041	.....	..... .0064
		Water added after heating	.....	.0016	.....	.0058	.....	.0085 .0082
27	Olaa Sugar Co., Ltd. Field 4-5	Immediately after treatment	.0043	.0040	.0062	.....	.....	.0045 .0086
		After 2 weeks	.0052	.....	.0056	.....	.....	.0043 .0065
91	Manoa Substation, surface, Field 7	Immediately after treatment	.0011	.0009	.0012	.....	.....	.0014 +.0100
		After 2 weeks	.....	.....	.0010	.....	.....	.0013 +.0100

Plus sign (+) denotes quantity greater than that indicated.

Since the soil cup is packed solidly, the regular R.C.M. tables giving ammoniacal and nitrate nitrogen values may be used, and correctly so, without further change.

The wire screen is washed and brushed and the excess water shaken off. It is then ready for use again without further drying.

#### CONCLUSIONS

A method for sampling moist field soils is proposed whereby reproducible analytical figures of available nitrogen (ammoniacal and nitrate) are obtainable quickly by means of rapid chemical methods. Such figures, based on the analysis of the original wet field soils are believed to be not only more representative of the actual condition of the field soil at the time of sampling, but are obtainable much more quickly and also more easily than when procedures are followed based upon regulation dried soil specimens.

Air drying or drying in the oven at 60° C. or 100° C., even for short periods, changed the available nitrogen content of soils to a great extent in most cases. From the available soil nitrogen viewpoint, the least objectionable method of soil drying is air drying.

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## Sunlight-Nitrogen Relationships

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By R. J. BORDEN

An abundance of sunlight and an adequate supply of available nitrogen are known to be prime assets for successful sugar production. Many of our field experiments have given results which attest to the effect of nitrogen upon both cane yields and its quality. Much less is actually known about the effects of sunlight or of the interaction of these two factors—sunlight and nitrogen. Consequently a skirmish test\* was undertaken to see what indication of this important relationship could be secured. The results of this preliminary test are now available (Table V), and although they are undoubtedly quite far from the complete answer, they are considered sufficiently interesting to present herewith for the discussion they may evoke and to stimulate still further investigation.

### TUE PLAN

To provide conditions that would be comparable and capable of being easily handled, the equipment of the Mitscherlich department was used. Seventy-two Mitscherlich pots were filled with identical amounts of thoroughly mixed Makiki soil, similarly fertilized with phosphate and potash, each planted with two uniform shoots of cane, and all given adequate irrigation without loss of drainage water during the whole period of growth. These pots were placed on flatcars which were run into the greenhouse during inclement weather and at night. To provide for the sunlight differentials that were introduced, one of the glass-roofed sheds was lined (roof and sides) with a single layer of natural burlap which provided a compartment in which the plants could be given a considerably reduced amount of direct sunlight; this was estimated by a Weston photronic exposure meter to be less than 5 per cent of the intensity of direct sunlight outside.

Two cane varieties were used: 31-1389 which starts off fast and grows very rapidly during its early life, and H 109 which starts more slowly and makes its greatest growth at a somewhat later stage.

### THE VARIABLES

Four "sunlight" variables were introduced during the latter part of the growing period while millable stalk was being formed. These may be identified as follows:

*Treatment A*—The controls. These plants were grown in all available direct sunlight from time of starting the crop on January 11, 1938 until it was harvested on March 29, 1939.

*Treatment B*—Decreased light *after* the "boom stage." These plants had all possible direct sunlight for the first 10 months (until November 30); thereafter for the four months from December 1 to March 28, they were given direct sunlight each morning only, being placed in the shaded house at noon each day.

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\* Experiment Station, H.S.P.A., Project A-105—No. 118.



*Treatment C*—Decreased light during the “boom stage.” These plants had all available direct sunlight for their first 6 months (until July 31); thereafter during the alternate months of August, October, December, and February, they received direct sunlight each morning only; during September, November, January, and March they again received all possible direct sunlight.

*Treatment D*—Intermittent light variation after the “boom stage.” Like *Treatment B*, these plants received direct sunlight daily for the first 10 months; thereafter they were left in the shaded house all day on Friday, Saturday, and Sunday of each week until harvested. They received full sunlight on other week days during this period.

Three nitrogen variables were included. The total amounts were divided into monthly applications and supplied during the first 9 months of growth:

*Treatment I* supplied each pot with a total of 5 grams of N which was considered an inadequate supply for a maximum yield.

*Treatment II* supplied the crop with 8.5 grams of N during the same period; this has been our standard practice for growing a good crop of cane in a Mitscherlich pot.

*Treatment III* furnished a total of 15.5 grams of nitrogen for the crop; this was undoubtedly more than could be efficiently used during the 14-month growing period.

#### OBSERVATIONS DURING GROWTH PERIOD

Certain pertinent observations were made after the differentials had been imposed:

- ✓ (1) There were very few live “tillers” in the pots which received an inadequate supply of nitrogen (I); although many suckers had started to grow in these pots, their early mortality had been exceptionally heavy.
- ✓ (2) An abundant and very vigorous sucker growth occurred in the pots which had received excessive nitrogen applications (III); similarly many of the upper eyes on the stalks had started to grow. (*Treatment II* was intermediate with I and III in its extent and character of tillering.)
- ✓ (3) Particularly striking was the effect of *Treatment C* upon the length of the internodes which were formed under its condition of reduced sunlight during August and October. These internodes were 2 to 3 times longer than respective internode growth made by the other plants which were in full sunlight during this period (Fig. 1). This effect of reduced light in increasing the internode elongation was not as prominent after December.
- (4) The internodes formed after December were distinctly shorter in the “controls” (*Treatment A*) than in plants which had not received the maximum of sunlight. However, the total number of millable joints was found to be slightly greater in these “controls.” Counts made shortly before harvest show the average number of dry-leaf millable joints for the nitrogen differentials to be quite similar, but for the sunlight differentials we have the following comparisons:

Treatment A—stalks average	36 dry-leaf joints
Treatment B—stalks average	33 dry-leaf joints
Treatment C—stalks average	31 dry-leaf joints
Treatment D—stalks average	30 dry-leaf joints



Fig. 1. Typical H 109 stalks showing the effects of reduced sunlight on elongation of internodes.

- A—Control—grown in full sunlight.
- C—Reduced light during the 7th, 9th, 11th, and 13th months.
- I—Supplied with inadequate nitrogen.
- II—Supplied with adequate nitrogen.
- III—Supplied with an excess of nitrogen.

## GENERAL EFFECTS

The cane was harvested at the age of 14 months. The data which were secured have been studied by analysis of variance to determine the significance of both of the main effects (nitrogen and sunlight) and of their interaction—separately for both varieties. Briefly this analysis may be summarized as follows:

Measurement	Variety	Effect of sunlight	Effect of nitrogen	Interaction of sunlight and nitrogen
Total dry weight	31-1389	Not significant	Significant	Not significant
Total dry weight	H 109	Significant	Significant	Not significant
Trash and tops only	31-1389	Not significant	Significant	Not significant
Trash and tops only	H 109	Not significant	Significant	Not significant
Millable cane	31-1389	Significant	Significant	Not significant
Millable cane	H 109	Not significant	Significant	Not significant
Quality ratio	31-1389	Significant	Significant	Not significant
Quality ratio	H 109	Not significant	Significant	Not significant
Sugar	31-1389	Significant	Significant	Not significant
Sugar	H 109	Not significant	Not significant	Not significant

From this summary we would be forced to conclude (a) that the response to length and period of direct sunlight exposure as given to these plants had been somewhat different for the two varieties; (b) that the generally expected effect of nitrogen had again been demonstrated; and (c) that there was no reliable indication of any interaction between sunlight and nitrogen, i.e., of any differential response to the nitrogen variables used with the several periods of direct and subdued sunlight conditions as they were imposed in this investigation.

## SUNLIGHT EFFECTS

More specifically, when we examine the significant data from Table I we note that the variety H 109 produced definitely more total dry matter when it was grown continuously in full sunlight, but that the reduced sunlight differences had no reliable effect upon its production of millable cane or its quality and final sugar yield. On the other hand, there was a pronounced tendency for the variety 31-1389 to make more millable cane and sugar when its exposure to direct sunlight had been somewhat restricted. Apparently also, a somewhat better cane quality was obtained from Treatment C in which the reduced sunlight was effective during the "boom stage."

TABLE I  
EFFECTS OF VARIATIONS IN SUNLIGHT  
(Averages from 9 Pots)

Sunlight treatments	Dry yield* per pot (gms.)		Dry weight of trash and tops only (gms.)		Wgt. millable cane per pot (lb)		Q.R.		In sugar	
	31-1389	H 109	31-1389	H 109	31-1389	H 109	31-1389	H 109	31-1389	H 109
A.....	1205	1320	483	523	3.86	3.77	7.10	6.78	.55	.56
B.....	1245	1209	483	499	4.47	3.48	7.50	6.91	.60	.51
C.....	1224	1240	453	491	4.54	3.83	6.84	6.56	.66	.58
D.....	1215	1202	472	491	4.55	3.54	7.31	6.73	.63	.53
Amount of difference needed for significance ..	62	73	41	38	.51	.57	.26	.31	.07	.09

\* Includes all trash, bagasse, solids in juice, and non-millable tops and leaves.

## NITROGEN EFFECTS

The data in Table II show that significantly lower total dry weights were secured from both varieties with the lowest amount of nitrogen supplied (Treatment I), and that there was no reliable increase for the excessive nitrogen application (III) over a moderate supply (II). A very definite increase in the amount of trash and cane tops was the result of increased nitrogen applications on both varieties.

With respect to the millable cane, the answer was different for the two varieties: the excess nitrogen application produced less millable 31-1389 cane than the lowest amount; but for best yields from H 109 the low amount was wholly inadequate.

The deleterious effect of increased nitrogen on cane quality is nicely shown: the heavy nitrogen applications (III) gave a cane with definitely poorer quality than the medium or low amounts.

TABLE II  
EFFECT OF DIFFERENT AMOUNTS OF NITROGEN  
(Average of 12 Pots)

Nitrogen treatments	Dry yield per pot (gms.)		Dry weight of trash and tops only (gms.)		Weight millable cane per pot (lb)		Q.R.		In sugar	
	31-1389	H 109	31-1389	H 109	31-1389	H 109	31-1389	H 109	31-1389	H 109
I.....	1127	1126	539	595	4.51	3.22	6.95	6.40	.65	.51
II.....	1253	1288	633	691	4.55	3.91	6.92	6.73	.66	.58
III.....	1286	1314	719	717	4.01	3.83	7.68	7.11	.52	.54
Amount of difference needed for significance ..	54	63	35	17	.44	.50	.24	.27	.06	.08

The final sugar yield from 31-1389 shows both the low and medium nitrogen applications to be superior to the high amount. With H 109, the sugar yields are not significantly different for the 3 nitrogen treatments and present-day economics would therefore indicate a preference for the lower amount.

## TRASH

A study of that percentage of the total dry weight which was made up by the trash and tops, shows that the variety 31-1389 was influenced to a greater extent than H 109, being affected more especially by the increases in nitrogen than by the variations in sunlight. Our earlier observations had indicated that the higher amount of nitrogen was producing a very abundant and vigorous sucker growth, which had not formed millable cane when the crop was harvested at the age of 14 months (Fig. 2). The variations in light were probably without definite effect on this trash: dry weight ratio with H 109 cane, but with 31-1389 we have, by inference, an indication that when the periods of reduced light had occurred all through the "boom stage" (Treatment C) the plants were more efficient in making millable stalk.

## DRY WEIGHT OF TRASH AND TOPS AS PER CENT OF TOTAL DRY WEIGHT

Sunlight treatment	31-1389	H 109	Nitrogen treatment	31-1389	H 109
A.....	53.4	52.8	I.....	47.8	52.9
B.....	51.7	52.3	II.....	50.5	53.6
C.....	49.4	52.8	III.....	55.9	54.6
D.....	51.7	54.5			



Fig. 2. Showing the effect of increased nitrogen applications upon the sucker growth.  
*I*—Inadequate Nitrogen; *II*—Adequate Nitrogen; *III*—Excess Nitrogen.

#### NITROGEN CONTENT

The percentage of nitrogen in the active green leaves at harvest may be related in some manner to the foregoing observations. The data in Table III show a higher percentage of total nitrogen in the leaves of plants which had received the larger nitrogen applications; this was especially so for the young sucker growth even though the greater part of this growth had been made after the last application of nitrogen, five months before harvesting.

TABLE III  
PER CENT TOTAL NITROGEN IN ACTIVE CANE LEAVES AT HARVEST

Variety	Sunlight treatment	Leaf sample* from	Nitrogen treatments			Sunlight treatment averages
			I	II	III	
31-1389	A	Primary stalks	.85	1.13	1.58	.92
		Suckers	.86	1.03	1.68	1.19
	B	Primary stalks	1.05	1.28	1.56	1.30
		Suckers	n.s.	1.11	1.40	1.26
	C	Primary stalks	.99	1.20	1.35	1.18
		Suckers	.73	1.05	1.56	1.11
	D	Primary stalks	.96	1.28	1.44	1.23
		Suckers	n.s.	1.29	1.80	1.55
	Nitrogen averages: Primaries		.96	1.22	1.23	
	Nitrogen averages: Suckers		.80	1.12	1.61	
H 109	A	Primary stalks	.90	1.07	1.69	1.22
		Suckers	.80	.99	1.56	1.12
	B	Primary stalks	1.20	1.35	1.46	1.34
		Suckers	1.04	1.20	1.54	1.26
	C	Primary stalks	.85	1.12	1.44	1.14
		Suckers	n.s.	1.28	1.70	1.49
	D	Primary stalks	1.06	1.44	....	1.25
		Suckers	.93	1.54	1.80	1.42
	Nitrogen averages: Primaries		1.00	1.25	1.53	
	Nitrogen averages: Suckers		.92	1.25	1.65	

\* Analysis from single leaf-punch sample only, but all stalks "punched" for same.  
n.s. = no sample.

The effect of variations in light upon the nitrogen content of the leaves of H 109 is perhaps not a significant one, but there is an indication that with 31-1389, the nitrogen applied had been more completely used in those plants which had been grown under the most abundant sunlight conditions. And this indication appears to be supported in a general way by the fact that the greater percentages of nitrogen in the primary stalks occur in Treatments B and D which did not get the full benefit of the sun during their last four months of growth.

The percentage of nitrogen in the crusher juice of both varieties was not affected significantly by the sunlight variations, but the increased nitrogen applications quite definitely increased the nitrogen content of this juice (Table IV).

TABLE IV  
EFFECT OF SUNLIGHT AND NITROGEN ON THE PER CENT NITROGEN  
OF CRUSHER JUICE

Treatments	Per cent nitrogen in crusher juice (averages of 3 pots)		
	31-1389		
	I	II	III
A.....	.033	.046	.158
B.....	.035	.061	.139
C.....	.029	.063	.148
D.....	.038	.050	.130
Treatments	H 109		
	I	II	III
	I	II	III
A.....	.029	.036	.051
B.....	.034	.038	.095
C.....	.030	.038	.078
D.....	.033	.040	.082

Amount of difference needed for significance between the nitrogen treatment averages:  
for 31-1389 = .023; for H 109 = .023.

## NITROGEN CONTENT AND CANE QUALITY

The harvest data indicate a rather broad, general relationship between the nitrogen content of either the leaf or the crusher juice, and the cane quality, i.e., when the treatment was such that it definitely increased the percentage nitrogen in the plant, a poorer quality resulted. This may be shown very nicely if we disregard the treatment differentials and use the complete set of data to determine the correlation and regression coefficients for nitrogen and quality ratio. Positive significant correlations have been obtained, as will be seen from the following:

Coefficient	Variety 31-1389	Variety H 109
1. Correlation (r), i.e., Percentage relation between		
(a) Q.R. and % N in leaf	$r = .72 \pm .14$	$r = .76 \pm .13$
(b) Q.R. and % N in juice	$r = .65 \pm .10$	$r = .63 \pm .10$
2. Regression, i.e., Estimate of Q.R.		
(a) from % N in leaf	$Q.R. = 5.56 + 1.35 \times \% N$	$Q.R. = 5.54 + .94 \times \% N$
(b) from % N in juice	$Q.R. = 6.69 + 6.57 \times \% N$	$Q.R. = 6.41 + 14.11 \times \% N$

TABLE V  
DETAILED HARVEST RESULTS  
(Averages of 3 pots, except as noted)

Variety	Treatment	Total dry weight (gms.)	Dry weight of trash and tops only (gms.)	Millable cane (lb)	Q.R.	Sugar (lb)	% total nitrogen in juice	% total nitrogen in (leaves* of) primary stalks suckers	
31-1389	A I	1072	553	3.82	7.1	.54	.033	.85	.86
	II	1260	654	4.00	6.6	.61	.046	1.13	1.03
	III	1283	724	3.77	7.6	.50	.158	1.58	1.68
	B I	1136	546	4.58	7.1	.65	.035	1.05	n.s.
	II	1267	650	4.75	7.3	.65	.061	1.28	1.11
	III	1333	735	4.08	8.0	.51	.139	1.56	1.40
	C I	1166	527	4.87	6.5	.72	.029	.99	.73
	II	1232	600	4.55	6.7	.68	.063	1.20	1.05
	III	1273	686	4.20	7.3	.57	.148	1.35	1.56
	D I	1136	529	4.77	7.1	.68	.038	.96	n.s.
	II	1251	627	4.91	7.1	.69	.050	1.28	1.29
	III	1256	730	3.97	7.7	.52	.130	1.44	1.80
H 109	A I	1168	643	2.93	6.3	.46	.029	.90	.80
	II	1319	702	3.95	6.8	.57	.036	1.07	.99
	III	1472	747	4.44	7.0	.63	.051	1.69	1.56
	B I	1019	552	2.51	6.5	.39	.034	1.20	1.04
	II	1293	705	4.15	6.9	.60	.038	1.35	1.20
	III	1314	738	3.78	7.3	.53	.095	1.46	1.54
	C I	1177	598	3.88	6.3	.61	.030	.85	n.s.
	II	1313	680	3.98	6.5	.61	.038	1.12	1.28
	III	1230	686	3.63	6.8	.53	.078	1.44	1.70
	D I	1138	589	3.57	6.3	.56	.033	1.06	.93
	II	1229	678	3.57	6.7	.54	.040	1.44	1.54
	III	1240	692	3.47	7.2	.48	.082	....	1.80

\* Simple composite sample only.

## CONCLUSIONS

1. The two cane varieties 31-1389 and H 109 responded somewhat differently to variations in exposure to direct sunlight. The effects were generally more significant upon 31-1389.
  2. The commonly accepted opinions concerning the various effects of nitrogen on cane growth and composition were quite nicely verified.
  3. Considered in the light of the technique that was used in this skirmish test, we were unable to show any significant interaction between sunlight and nitrogen.
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## Sugar Prices

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96° CENTRIFUGALS FOR THE PERIOD  
MARCH 16, 1939 TO JUNE 14, 1939

Date	Per pound	Per ton	Remarks
Mar. 16, 1939..	2.85¢	\$57.00	Puerto Ricos.
“ 18.....	2.84	56.80	Puerto Ricos, 2.83; Philippines, 2.85.
“ 20.....	2.85	57.00	Cubas.
“ 21.....	2.87	57.40	Philippines.
“ 28.....	2.86	57.20	Philippines, Puerto Ricos.
“ 29.....	2.88	57.60	Puerto Ricos.
Apr. 5.....	2.90	58.00	Puerto Ricos, Philippines.
“ 12.....	2.88	57.60	Cubas.
“ 13.....	2.92	58.40	Philippines, Puerto Ricos.
“ 14.....	2.935	58.70	Cubas, 2.95; Puerto Ricos, 2.92.
“ 18.....	2.95	59.00	Puerto Ricos.
“ 21.....	2.93	58.60	Cubas.
“ 28.....	2.89	57.80	Cubas.
May 8.....	2.925	58.50	Puerto Ricos, 2.92; Philippines, 2.93.
“ 9.....	2.915	58.30	Puerto Ricos, 2.93; Cubas, 2.90.
“ 10.....	2.94	58.80	Philippines.
“ 11.....	2.95	59.00	Philippines.
“ 16.....	2.90	58.00	Cubas.
June 2.....	2.86	57.20	Philippines.
“ 6.....	2.87	57.40	Puerto Ricos.
“ 8.....	2.85	57.00	Cubas.
“ 14.....	2.80	56.00	Puerto Ricos.

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# THE HAWAIIAN PLANTERS' RECORD

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## **In This Issue:**

### *The Cane Variety 31-1389:*

Germinated in 1931, the variety 31-1389 has been extended rapidly. It now occupies nearly twenty thousand acres. Although it is already being superseded by newer seedlings in some districts, it still commands interest in the districts to which it is best suited. The characteristics of this variety are examined in a series of five papers which deal with its origin, its adaptability to various climatic conditions, its reaction to diseases, its susceptibility to insect attack, its response to fertilizers, and its manufacturing qualities.

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### *A Lysimeter Study of Losses of Applied Potash:*

Potash applied to an acid soil in lysimeters suffered insignificant losses by leaching when the pots were cropped to cane. Uncropped pots suffered considerable losses through leaching. Ratoon cane appears to exhibit a high degree of efficiency in utilizing applied potash fertilizers.

The effective base-exchange capacity of the soil seems to play a very minor role compared with that of the cane crop in conserving applications of potash in fertilizers.

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### *Disease Control and Stimulation of Cane Cuttings by the Hot-Water Treatment:*

The subject matter in this article deals with the development of the hot-water treatment in relation to the control of chlorotic streak disease and the application of the treatment as a plantation practice for disease control as well as for stimulating the germination of sugar cane cuttings.

*Evaporation of Moisture from Soil in Large Lysimeter Pots:*

An experiment which required measurement of water applied and water leached from uncropped lysimeter pots indicated considerable losses of water by evaporation. These results are reported without comment.

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## 31-1389—Its Origin and Present Status

By A. J. MANGELSDORF

### INTRODUCTION

Even the best of our present varieties of sugar cane fall short of perfection. The perfect cane may be beyond our imagination, but we can set down some of the specifications for an improved variety in terms of the varieties with which we are familiar. Our hypothetical variety might, for example, combine such characteristics as:

1. The light tasseling, clean stalk, good milling qualities and eye spot resistance of Yellow Caledonia, without its weak ratooning.
2. The second-season growing power and high juice purity of H 109 without its eye spot susceptibility and slow starting.
3. The ratooning and weed suppressing power of POJ 36 without its slenderness and low sucrose content.
4. The high sucrose content of POJ 2878 without its boiling-house difficulties and excessive tasseling.

Such a list of specifications, while far from complete, serves to illustrate the fact that the desirable qualities are widely scattered among our present varieties. The objective of sugar cane breeding is to synthesize new canes which combine a maximum of the desirable qualities with a minimum of the undesirable qualities.

31-1389 may be regarded as one of the steps toward this objective. The purpose of this discussion is to record the history of 31-1389 and to attempt an evaluation of its desirable qualities and its faults, together with a consideration of their bearing upon its suitability for our various districts.

### PROPAGATION AND SELECTION OF 31-1389

31-1389 is the result of a cross between POJ 2878 and 26 C 270. The cross was set up at Kailua substation on November 29, 1930, under the supervision of J. N. P. Webster.

Because of its world-wide reputation there was naturally a strong interest in POJ 2878 as a breeding cane. The 1930-1931 crossing season gave us our first tassels of the "Java supercane." The lack of information as to the reaction of its tassels to the  $\text{SO}_2$  solution led us to undertake a number of field crosses in which the POJ 2878 tassels used as females were allowed to remain growing on their own roots.

Tasseling stalks of the local varieties which were to serve as the male parents were cut and placed in the standard  $\text{SO}_2$  solution. These cut tassels were then carried to the tasseling stools of POJ 2878 and supported in position so that their pollen would fall upon the tassels of POJ 2878.

The cross which produced 31-1389 was carried out in this manner. The tassels were harvested when ripe and germinated in the Makiki greenhouse. The germinating flat yielded 130 seedlings, which were set out in Makiki, Field 17, during April 1931 along with the seedlings from many other crosses.

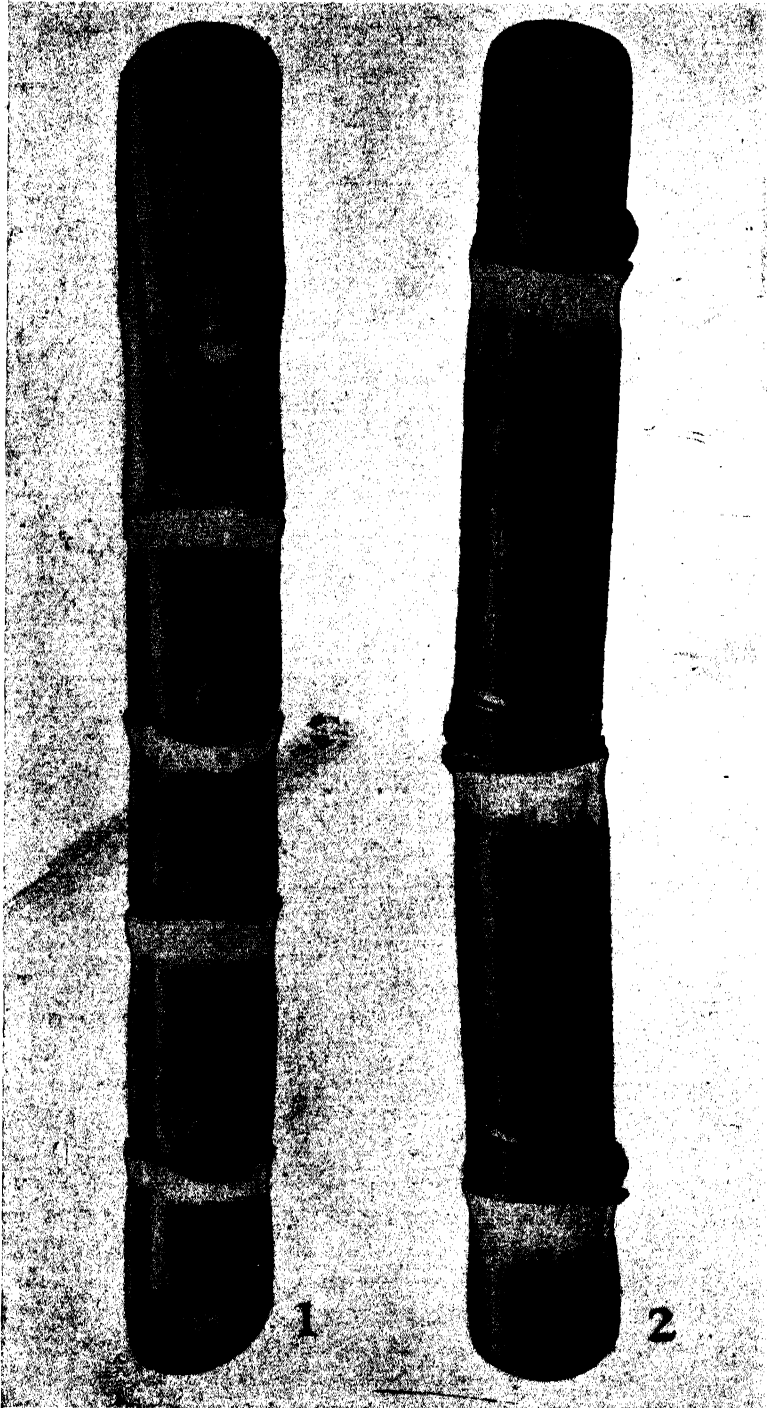


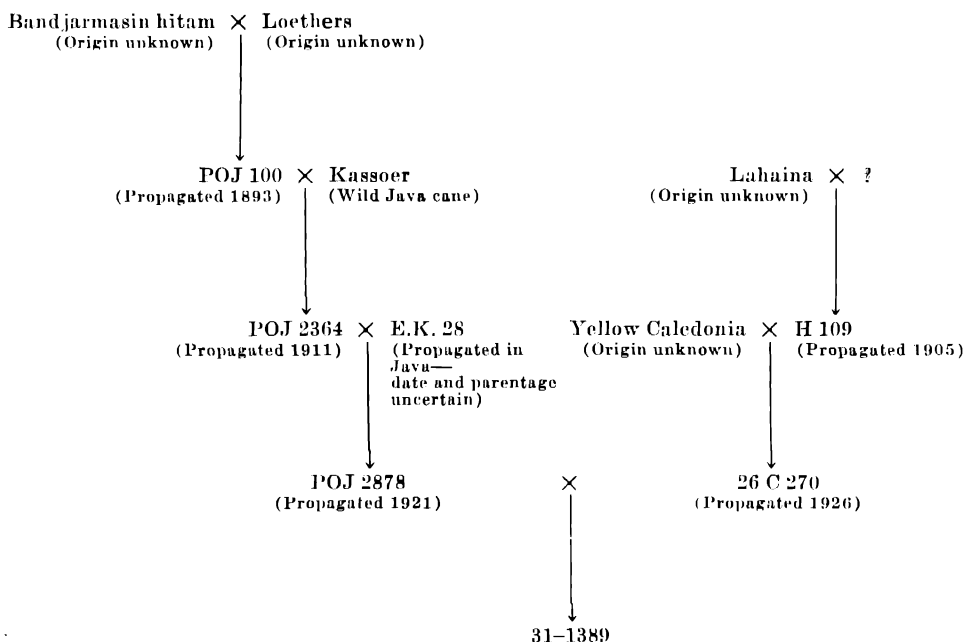
Fig. 1. Seedpieces of 31-1389. Front and side views showing extent of wax band.

The first selection of this group of seedlings was made by C. G. Lennox on September 28, 1931. The seedling which was to receive the number 31-1389 had six stalks, and a Brix of 14.5, which was above average for this field of young cane. It was given a grade of plus and was sent to the Kailua and Waipio stations for further testing.

The ratoon selection of this field was made on March 18, 1932, at which time the ratoon stool of 31-1389 was noted as double plus, with 17 stalks and a Brix of 15.8, which again was above average. J. A. Verret, Consulting Agriculturist of this Station, visited the field from time to time during the course of the selection. When Mr. Verret saw this stool he was so favorably impressed that he requested W. Twigg-Smith to make a photographic record of it. The photograph is reproduced on the cover of this issue.

The seed from the ratoon stool was sent to the Waipio, Kailua and Manoa stations for further testing and the extra body seed was sent to Kailua for spreading.

#### PEDIGREE OF 31-1389



The pedigree of 31-1389 reveals its distinguished ancestry. Its mother and its maternal grandfather (E. K. 28) were the leading varieties of Java in their time. 26 C 270, the father of 31-1389, is the descendant of the three outstanding canes in Hawaiian sugar history. Lahaina was our leading variety until its failure during the early years of this century when it gave way to Yellow Caledonia, which held first place until 1924. In that year the Lahaina dynasty regained its leadership through the rise of H 109.

However, even the most distinguished of the ancestors of 31-1389 have their shortcomings, and some of its forbears are definitely mediocre by present standards. Unfortunately, 31-1389 has inherited some of the faults of its ancestors, along with many of their virtues.

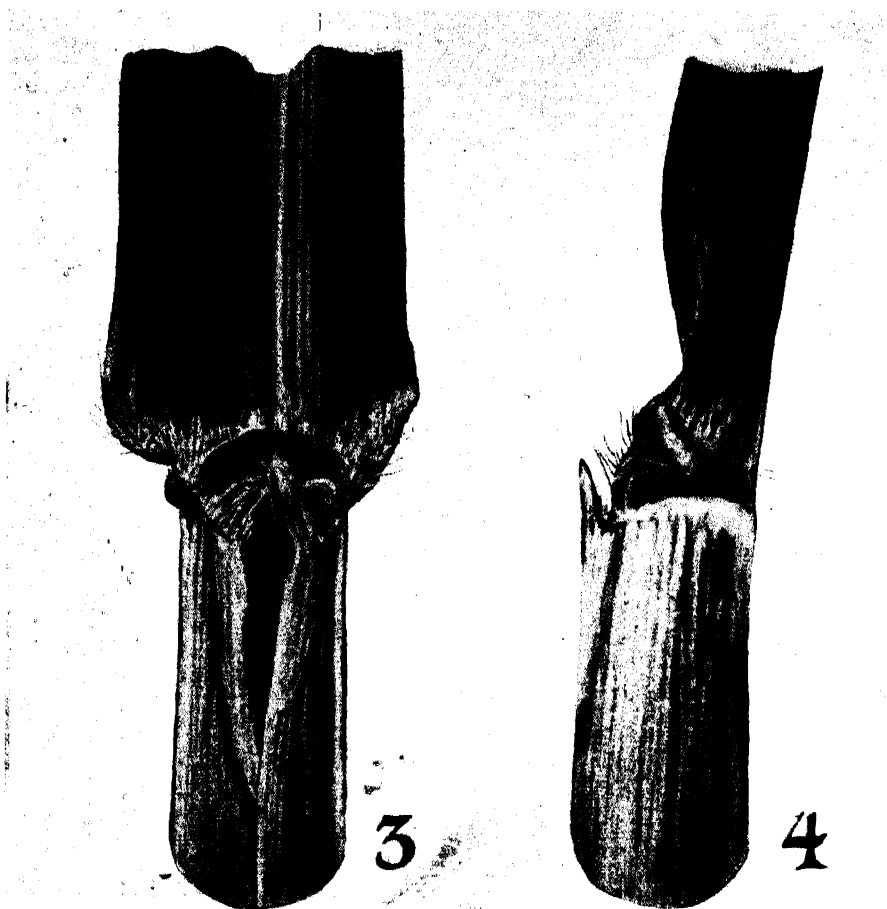


Fig. 2. Collar of 31-1389. Front and side views showing auricle and pubescence behind ligule.

#### DESCRIPTION OF 31-1389

Slender at base, increasing in diameter in older cane, especially during late winter and spring. Average diameter larger than POJ 2878 and slightly smaller than H 109.

Rind very soft as young cane but tends to harden with maturity. Softer than H 109 but slightly harder than POJ 2878.

Color of young (unexposed) internodes, pale greenish-yellow.

Color of exposed internodes, yellow to brownish-yellow, sometimes tinged with green or russet.

Wax band narrow, conspicuous, more sharply defined than in POJ 2878 (Fig. 1).

Flesh color, as seen in cross section of internode, pale brownish-green or olive-green at rind, fading to nearly white at center.

Eyes plump, round, with well-defined wings. Hairing at apex of eye inconspicuous or absent as contrasted with the eyes of POJ 2878, the tips of which carry a tuft of hairs. Eye-groove absent (Fig. 3).

Root primordia usually three or four rows near eye, decreasing to two rows on opposite side of stalk.

Leaves pale green, often freckled, blades tending to curl backward toward midrib.

Hairing on back of leaf sheath (Group 57). Hairs fairly stiff and sharp but more or less appressed against leaf sheath. Hairing less formidable and much less extensive than on POJ 2878.

Joint triangle (dewlap) large, conspicuous, reddish in the younger leaves, inner surface downy pubescent. Auricle well developed on underlying edge of leaf sheath, absent on overlapping edge (Fig. 2).

#### CULTURAL CHARACTERISTICS OF 31-1389

*Germination:* The round prominent eyes of 31-1389 are easily damaged in handling. This is especially true of body seed from which the protecting leaf

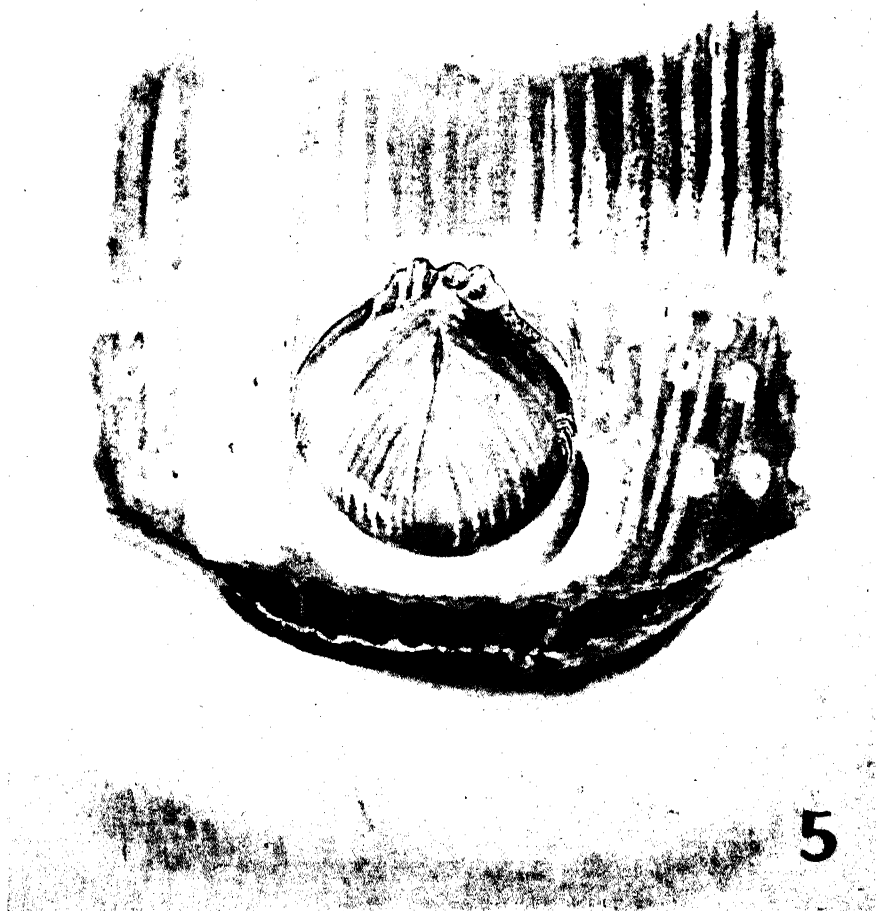


Fig. 3. Node of 31-1389 (enlarged) showing relative positions of leaf scar, eye and root primordia.



sheaths have been removed. Poor germination has frequently been experienced from stripped body seed handled in bags.

Several plantations have reported a reduction in damage to eyes when body seed is handled in bundles.

Unstripped top seed can be handled without special precaution, since the eyes are well protected by the overlying leaf sheaths.

*Ratooning:* As a fast starter in ratoons 31-1389 is one of the best. However, there is some difference of opinion as to its ability to maintain yields in ratoons. In some cases ratoon yields have been disappointing—in other cases yields have improved in ratoons.

Lack of soil aeration often limits ratoon yields in tight, poorly drained soils and in soils which pack badly. In such soils the plant crop is benefited by the improvement in aeration which results from plowing. However, the heavy soils tends to relapse quickly to a compact condition as a result of irrigation and of packing by heavy equipment, especially if harvested in wet weather. Under such conditions the ratoon yields of all varieties are likely to decline badly as compared with the plant crop. 31-1389 is no exception to this rule.

We have tallied the data from 62 replicated tests for which both plant crop and first ratoon data are available.

In 30 of the 62 tests 31-1389 improved its relative standing in first ratoons as compared with its plant crop standing. In 31 tests it compared less favorably in first ratoons than in plant. In one test there was no change in its relative standing in plant and first ratoons.

From the above it appears that 31-1389 is on the whole neither better nor worse in ability to maintain its yield in first ratoons than the standard varieties with which it was compared.

Wailuku reports that unlike POJ 2878, 31-1389 maintains a good stalk diameter in ratoons.

*Closing-in:* 31-1389 is outstanding as a fast starter. It develops its secondary shoots early and in abundance. Few varieties can equal it for fast closing-in. This is especially true under the makai conditions to which it is best suited.

As young cane it has an erect habit which together with its curving leaves makes it an excellent variety for mechanical cultivation.

Even in situations like the upper lands of the Hilo Coast and of Windward Kauai, to which it is not well suited, its early growth is rapid. Under these conditions, however, the stalk population is sparse and the crop tends to become open after going down.

*Reaction to Environment:* 31-1389 is at its best in the porous, fluffy, residual soils, and in the well-drained, gravelly alluvial soils. When grown on tight, poorly drained, and poorly aerated soils it may be benefited by sub-soiling, if the operation can be done under dry conditions immediately after harvesting.

31-1389 is inferior to H 109 in salt tolerance. At Ewa, at Waianae, and in the Kekaha flats, where the irrigation water is saline it has failed to outyield H 109. Its characteristic leaf freckle is aggravated under these conditions, which suggests that the freckle may be caused by an excessive intake of one or more of the salts which occur in abundance in such soils, or by a lack of availability of some essential element under the prevailing alkaline conditions.

Its drought resistance is better than average. Under severe drought it has shown more windburn than POJ 2878, but it makes a faster recovery after the moisture supply has been replenished. In the drought-resistance tests at the Waipio and Kailua substations it is among the leaders.

Its leaves are easily shredded by wind, but it is nevertheless a strong competitor in such windy situations as the makai lands of Hamakua, Kohala, Olowalu, and Central Maui.

31-1389 is typically a makai cane. Under mauka conditions it is usually pale and anaemic in appearance. In spite of its poor appearance it has occasionally given a good account of itself under middle-belt and mauka conditions. It has yielded well at 1000 feet in the Hamakua District and is a strong competitor in the mauka lands of Oahu Sugar Company.

*Tasseling:* 31-1389 may be classed as a medium tasseler. Counts from adjoining plots have shown lower tasseling percentages for 31-1389 than for POJ 2878.

The following figures are typical:

Kaeleku Sugar Company, Ltd., age 8 months.		
	No. counted	Per cent tasseled
31-1389 .....	500	4
POJ 2878 .....	500	27
31-1389 .....	1000	18
POJ 2878 .....	1000	41

As mature cane, 31-1389 usually tassels somewhat less than adjoining H 109 of the same age. However, when started in April or May, H 109 because of its slowness in the early stages may still be too small to tassel in November and December, while 31-1389 is likely to produce an occasional tassel.

Under sunny makai conditions the tasseled stalks of 31-1389 develop strong lasas which contribute materially to the tonnage at harvest. Under cloudy windward mauka conditions the lasas are slower and the weaker stalks may die through failure to develop lasas. The pithiness which develops after tasseling is more extensive than in H 109 but less than in POJ 2878.

*Harvesting Characteristics:* Because of its good stalk size, soft rind, and easy stripping 31-1389 is a desirable cane for harvesting by hand. For mechanical harvesting it has the advantage of good anchorage, the stools being less inclined to uproot than H 109, thus requiring less replant in the following crop.

McBryde reports that 31-1389 involves more pickup after rake harvesting since the stalks tend to break off several feet above the base. It is felt, however, that this fault is less costly than the uprooting and consequent heavy replant encountered in H 109.

*Optimum Crop Length:* The growth curve of 31-1389 differs from that of H 109. 31-1389 grows rapidly during the first season and slows down during the second season. H 109 is slow in getting under way, but it gains momentum during the second season. Under comparable conditions 31-1389 will attain in 18 to 20 months the tonnage and maturity characteristic of H 109 at 22 to 24 months.

The rind of 31-1389 is softer than that of H 109 and the stalks are more liable to injury and deterioration. 31-1389 is attractive to rats, and is likely to suffer serious damage in the presence of a large rat population. All of these features

point to shorter cropping cycles for 31-1389 than for canes like H 109 and Yellow Caledonia.

An examination of the data from replicated tests shows that 31-1389 is fairly flexible in respect to crop length. At Lihue it has given good results when harvested at 12 months. At Hawaiian Commercial and Sugar it held its own with H 109 at 26 months. In general, however, it may be said that 31-1389 may be successfully carried beyond 20 months only in the dry, sunny, well-drained situations. In the moist cloudy districts it must be harvested at 18 months or less for best results.

*Juice Quality:* Forty-nine replicated tests have been harvested to date comparing 31-1389 with H 109.

The average juice figures from these tests are tabulated below:

	Brix	Pol	Purity	Y% C	Q.R.
31-1389 ....	18.5	15.5	83.9	11.30	8.95
H 109 .....	17.9	15.4	86.1	11.46	8.73

It will be noted that 31-1389 averages higher in Brix and lower in purity than H 109 with little difference in the average quality ratios of the two canes. In this respect 31-1389 resembles its mother, POJ 2878, which shows a similar Brix-purity relationship.

It is encouraging to note that 31-1389 was nearly equal to H 109 in average quality ratio in spite of the fact that the tests were carried out under the fertilizer practices and crop lengths which have been adopted for H 109. It is reasonable to expect that an improvement in the juice quality of 31-1389 will result from a better understanding of its requirements.

The juice quality of 31-1389 is not equal to that of POJ 2878 when the Java cane is at its best, but when POJ 2878 has produced a heavy crop of suckers after tasseling its juices are frequently poorer than those of 31-1389. On the whole 31-1389 is average in juice quality. Very good or very poor juices are seldom encountered.

#### DISTRIBUTION OF 31-1389

31-1389 made its appearance just as the first regional variety stations were getting under way. Prior to that time the absence of regional stations necessitated sending new seedlings of promise to the plantations for their preliminary testing. During the transition period this practice was continued to the extent of distributing to the plantations seedlings which appeared outstanding without waiting for what we now regard as essential data from preliminary tests at the regional stations.

Such was the case with 31-1389. Its remarkable growth at Makiki, Kailua, and Waipio attracted immediate attention. In 1932, less than two years after germination, it was distributed to four plantations for further study. In 1933, it was sent to 24 plantations. Its appearance in these early plantings was so encouraging that by March 1934, within three years of the date on which it was transplanted from a pot into the field, it had been distributed to every plantation in the Territory.

Honolulu Plantation Company was the first to plant 31-1389 in a replicated variety test, and it was also the first to spread the new cane to field-scale plantings. H 109 and POJ 2878 were the leading varieties on this plantation at the time. Neither of these canes had proved entirely satisfactory. The slowness of H 109 as young cane resulted in high weeding costs. POJ 2878 gave good returns in the plant crop but declined in ratoons. The management was keenly interested in find-

ing a better variety for the plantation and pursued a vigorous testing program with this object in view. By the end of 1934, 31-1389 had been spread to 200 acres at Aiea. In 1935, 807 acres were planted and in 1936, 401 acres. By 1937 the area of 31-1389 at Honolulu Plantation Company had reached a total of 1777 acres.

During this time 31-1389 was being tested and spread to field areas on other plantations as well. In 1934, 6 replicated tests were installed to compare the new cane with the standard varieties. In 1935, 27 tests were installed and in 1936, 39 tests.

The increase in the acreage of 31-1389 on each of the four Islands from 1931 to 1938 is shown in the following table:

	Acreage of 31-1389 (1933 to 1938)					
	1933	1934	1935	1936	1937	1938
Kauai .....	0	60	763	2036	2745	3784
Oahu .....	1	286	1641	2628	4161	6325
Maui .....	0	92	493	1573	2490	3169
Hawaii .....	0	0	61	452	1757	3633
Total for Territory	1	438	2958	6689	11,153	16,911

It is of interest to compare the rate of increase of 31-1389 with that of H 109. Germinated in 1905, H 109 first appears in the acreage census of 1914, with 26 acres. In eight years 31-1389 has reached an acreage as large as that attained by H 109 in fifteen years.

The acreage of 31-1389 as of December 31, 1938 is shown below. Only plantations with one hundred acres or more are listed.

Plantation	Acreage of 31-1389 as of December 31, 1938	Per cent of total cane area on plantation
Kilauea .....	560	15.4
Lihue .....	1553	10.8
Grove Farm .....	433	13.0
McBryde .....	825	16.0
Kekaha .....	282	4.0
Honolulu Plantation Co...	1752	34.9
Oahu Sugar Co.....	824	7.3
Waialua .....	1293	13.1
Kahuku .....	895	19.5
Waimanalo .....	1528	56.1
Pioneer .....	998	10.0
Wailuku .....	1147	25.8
H. C. & S. Co.....	218	1.6
M. A. Co.....	309	3.7
Kaeleku .....	497	15.0
Kohala .....	1110	9.2
Paauhau .....	318	7.3
Kaiwiki .....	121	2.9
Hakalau .....	149	2.5
Honomu .....	883	29.2
Pepeekeo .....	483	12.7
Hilo .....	134	1.7
Hutchinson .....	100	2.0
Total for Territory.....	16,911	7.2

## PERFORMANCE OF 31-1389

One hundred and five replicated tests comparing 31-1389 with other varieties have been harvested by the plantations to date. These have been reported in detail in the Director's Monthly Report and the figures will not be repeated here.

In considering the results it is well to remember that even tests which are well replicated and carefully harvested are subject to error if situated on variable land. We cannot hope to escape soil variability entirely, nor can we hope to escape entirely errors in harvesting and in juice sampling. In view of these sources of error, and in further view of the climatic differences from season to season, we shall have to expect occasional contradictions. At best the replicated test is a crude tool, but since we know of no better one it devolves upon us to use it intelligently and to safeguard it to the best of our ability against unnecessary error.

With the limitations of replicated tests in mind, we may proceed to a consideration of the results of the tests comparing 31-1389 with other canes.

**KAUAI:**

*Kilauea*—In seven harvestings of replicated tests 31-1389 was better than POJ 2878 four times, it was equal twice, and poorer once. 31-1389 has a decided advantage over POJ 2878 in fields which are subject to eye spot and in cycles which favor tasseling.

31-1389 does well at Kilauea only on the better makai lands. Under the poorer conditions it has been outyielded by 31-1322 and by 31-2447. Even under makai conditions it has recently lost to 31-2806 in one test and to 32-8560 in two tests. It seems doubtful whether 31-1389 can hold its own against these newer canes even under the best of Kilauea conditions.

*Lihue*—31-1389 has not distinguished itself in replicated tests at Lihue. It has lost in several tests to 28-2055 in the middle and upper lands, and 28-2055 has in turn been outyielded by 31-2806, 32-1063, and 32-8560. 31-1389 has also lost to 31-2482, 31-2510, 31-3243, and 32-8163 in one or more tests. It is obviously out of the running in the poorer, middle-belt and mauka lands.

Its field performance in the better lands, however, has been quite satisfactory. In several fields it has established new records in sugar per-acre-month.

*Grove Farm*—31-1389 has not been included in replicated tests on this plantation, but it has yielded well in field plantings on the better makai lands.

*Kipu*—31-1389 has not been tested in replicated experiments on this plantation. Field observations indicate that it is not suited to Kipu conditions.

*McBryde*—31-1389 appears in only one test where it has outyielded H 109 and has equalled POJ 2878. It is preferred to H 109 because of its faster closing-in and lower cultivation costs.

*Kekaha*—In the makai lands 31-1389 has lost repeatedly to 28-2055 and 28-3540. Judging from its performance under similar conditions elsewhere it may still have possibilities in the lower half of the mauka slopes.

**OAHU:**

*Honolulu Plantation Company*—As already mentioned this plantation was the first to adopt 31-1389 as a commercial cane. H 109 had been for many years the standard variety but it was not at its best under Aiea conditions. The superiority of 31-1389 from the standpoint of cultivation costs was obvious from the start and

no time was lost in spreading it to field-scale plantings. 31-1389 has since lost to 32-8560 in both plant and ratoons of a replicated test and the results of field harvestings also point to the superiority of the latter cane. 31-1389 may eventually be forced to give way to 32-8560 at Aiea.

*Oahu Sugar Company*—31-1389 has been compared with H 109 in 12 harvestings of replicated tests (including plant and ratoons). In five of these it outyielded H 109, in five the yields were approximately even, and in two harvestings H 109 outyielded 31-1389. In field performance 31-1389 has shown up to best advantage in the upper lands where the slow growth of H 109 is further aggravated by eye spot disease. In tests recently harvested 31-1389 has twice lost to 32-8560, once in the middle belt and again in a makai "island" field.

*Ewa*—In six harvestings, H 109 led four times, while 31-1389 barely equalled H 109 in the other two. 31-1389 is not regarded with favor at Ewa. This applies also to Waianae, where the conditions are similar.

*Waialua*—In nine harvestings of replicated tests (plant and ratoons) 31-1389 led H 109 five times, the two canes were practically even three times, while H 109 led only once. No tests have as yet been harvested at Waialua comparing 31-1389 with 32-8560, but from observation the latter cane is believed to be superior in yielding ability and is being spread in preference to 31-1389.

*Kahuku*—Seven harvestings comparing 31-1389 with H 109 have resulted in substantial gains for 31-1389 in every case. The superiority of 31-1389 over H 109 at Kahuku in weed control and in ratooning ability is striking.

*Waimanalo*—POJ 2878 has run a close race with 31-1389 in replicated tests, most of which, however, were harvested in a non-tasseling cycle favorable to POJ 2878. Field experience has shown that 31-1389 involves less risk in a heavy tasseling cycle. This is especially true in fields which suffer from eye spot disease. 31-1389 has replaced POJ 2878 in the majority of the Waimanalo fields.

#### **MAUI:**

*Pioneer Mill Company*—31-1389 has been tested chiefly against POJ 2878 and 28-2055. In eleven harvestings comparing POJ 2878 with 31-1389, the latter cane led six times, POJ 2878 led three times, twice their yields were even.

In ten harvestings comparing 31-1389 with 28-2055 at Pioneer, four favor 31-1389, two favor 28-2055, and four were even. 31-1389 is being used extensively in the Olowalu Section. Whether or not it will be able to establish itself in the mauka lands above the H 109 belt will depend upon its ability to compete with 32-8560 and 32-1063 under these conditions.

*Wailuku*—31-1389 has appeared in only two replicated tests, one against H 109 and the other against POJ 2878. In neither test was 31-1389 definitely superior. Except for its larger stalk it has little advantage over POJ 2878 in a non-tasseling cycle, but for harvesting during the first half of the year it involves less risk of heavy tasseling in the following crop. Because of this and because of its low cultivating costs as compared with H 109 it has been spread to over one-fourth of the total area at Wailuku.

*H. C. & S. Co.*—31-1389 has outyielded H 109 in two tests, but with low odds. It is more tolerant of the growth-failure spots than H 109, and is being spread in areas where H 109 is not at its best.

*M. A. Co.*—31-1389 has been included in four tests, only one of which showed a definite gain for 31-1389. In two tests it barely equalled H 109 and in one test in the Hamakuapoko Section it lost by a small margin to POJ 2878. It is considered approximately equal to H 109 in yielding ability and superior from the standpoint of cultivating costs.

*Kaeleku*—31-1389 has led POJ 2878 in three out of five harvestings. Here again 31-1389 has little advantage over POJ 2878 except in a cycle which permits the Java cane to tassel heavily.

#### **HAWAII:**

*Kohala*—Two out of seven harvestings favored POJ 2878, two favored 31-1389, and three were even. In the middle-belt, unirrigated lands both 31-1389 and POJ 2878 lost to 32-3575 by two tons of sugar. 31-1389 has been spread to 1100 acres in the makai irrigated lands as an alternate for POJ 2878, which already occupied 3000 acres on this plantation.

*Honokaa*—31-1389 equalled POJ 2878 in a single test. It is favorably regarded as a cane for the makai irrigated lands.

*Paauhau*—There have been no direct comparisons of 31-1389 and POJ 2878 in replicated tests at Paauhau, but 31-1389 is preferred because of its good field performance in the irrigated lands. However, 31-1389 may have to give way to 32-8560, which outyielded it by a large margin in a recently harvested preliminary test.

*Hamakua Mill Company*—31-1389 outyielded POJ 2878 in two out of three harvestings, the third showing no difference.

*Kaiwiki Sugar Company*—Seven harvestings have shown no consistent difference in the yielding ability of 31-1389 and POJ 2878.

*Laupahoe*—31-1389 outyielded Yellow Caledonia by a good margin in a makai test. In the high rainfall district, from Laupahoe to Olaa, 31-1389 must be harvested at 18 months of age or less to avoid the risk of excessive deterioration.

*Hakalau*—In two out of four harvestings 31-1389 came through with good gains over POJ 2878. The other two showed small gains for POJ 2878. Both canes have been outyielded by 31-2510. The latter cane merits further study as a short cropper for the makai lands of the Hilo Coast.

*Honouliuli*—31-1389 has been compared with POJ 2878 in a single test. In the plant crop POJ 2878 led by .7 TS/A, in first ratoons 31-1389 led by 1.0 TS/A. 31-1389 is preferred on the basis of its field performance under short cropping in the makai lands.

*Pepee*—31-1389 outyielded Yellow Caledonia in both plant and first ratoons of a makai test. Its field performance under short cropping has been good.

*Onomea*—31-1389 was badly beaten by POJ 36 in a middle-belt test harvested at 25 months. Its soft stalk and recumbent habit disqualify it for long cropping in this wet district.

*Hilo Sugar*—In a test at 1100 feet 31-1389 led POJ 2878 in plant and lost in first ratoons. Neither cane can measure up to 32-1063 or 31-2484 under these mauka conditions.

*Olaa*—In one test in the Kapoho Section, 31-1389 outyielded Yellow Caledonia. In another test in the Paho Section it equalled Olaa 3117, but lost to 31-2207.

*Hawaiian Agricultural Company*—In the single test harvested thus far 31-1389 outyielded D 1135 but lost to UD 50. The test is situated in the middle-belt. According to present indications neither cane will be able to compete with 32-1063 and 32-8560.

*Hutchinson*—31-1389 lost to POJ 2878 in two out of six harvestings and barely held its own in the other four. Here again it seems likely that both canes will have to give way to 32-8560 which outyielded POJ 2878 by a large margin in a recently harvested test.

#### 31-1389 AS A BREEDING CANE

During the past four years 31-1389 has been used extensively in crossing with the object of combining its desirable qualities with those of certain other canes. It has been crossed with seedlings of Uba and *spontaneum* derivation to incorporate tolerance for mauka conditions and with *robustum* derivatives to develop a harder rind.

Even our best breeding canes produce only a small percentage of seedlings sufficiently free of faults to warrant further testing. This holds true for crosses with 31-1389. However, the general level of the 31-1389 crosses is above average, and some of the 31-1389 seedlings now in the preliminary stages of testing appear to have commercial possibilities. A few are fully equal to 31-1389 in fast starting and abundant stooling. 31-1389 promises to be a useful stepping stone in sugar cane breeding.

#### SUMMARY

The principal faults of 31-1389 are:

- (1) A soft rind which results in deterioration under long cropping. This difficulty is aggravated by wet conditions.
- (2) Poor fuel value of bagasse.
- (3) Sensitiveness to poor drainage and lack of soil aeration.
- (4) Sensitiveness to poor mauka soils, especially on the windward slopes.
- (5) A prominent eye which is easily damaged in handling when exposed by removing the leaf sheath.

The desirable features of 31-1389 are:

- (1) Fast starting, excellent stooling, good closing-in and strong ratooning, all of which should help to reduce cultivation costs.
- (2) Good drought resistance, which should make for more effective use of irrigation water.
- (3) A relatively low nitrogen requirement, which should result in reduced fertilizer costs.
- (4) Resistance to eye spot disease and mosaic disease.
- (5) Moderate tasseling.
- (6) Easy cutting when harvested by hand.
- (7) Strong anchorage for grab or rake harvesting.

Present status and future prospects of 31-1389:

- (1) 31-1389 is not suited to the tight, saline soils of Kekaha, Ewa, and Waianae.
- (2) It is not suited to the poor mauka lands of the unirrigated districts.
- (3) It cannot hold up under long cropping in the Hilo District.
- (4) It is barely equal to POJ 2878 as a short crop cane on a non-tasseling cycle.



(5) It is equal to or better than H 109:

- (a) in the makai lands of Kilauea, Lihue, and Grove Farm;
- (b) on residual slopes like those of McBryde, Aiea, Kahuku, and Wai-luku;
- (c) on alluvial soils like those of Olowalu;
- (d) in the poorer fields of Central Maui;
- (e) in fields where H 109 suffers from eye spot;
- (f) under conditions of labor shortage, since 31-1389 suffers less than H 109 from unavoidable delay in weeding and irrigation.

(6) It is equal to or better than POJ 2878 in the makai irrigated lands of the Kohala and Hamakua districts.

(7) It has produced good yields in sugar per-acre-month as a short cropper in the makai lands of the Hilo District.

31-1389 was gaining rapidly in acreage in the districts to which it is suited until the advent of new canes like 32-8560. The latter cane is not equal to 31-1389 in the early stages, but the preliminary data have shown it to be a heavier yielder at harvest.

Many replicated tests comparing 31-1389 with the more recent canes are already under way. From the harvesting results it will be possible within the next year or two to form a more accurate opinion as to the future of 31-1389.

### 31-1389—Its Reaction to Cane Diseases

By J. P. MARTIN

Within the past five years the variety 31-1389 has been commercially grown under a wide range of environmental conditions throughout all the Islands and has been exposed in one locality or another to natural conditions most favorable for the development of each of the major diseases. During this period numerous field observations have been recorded regarding its reaction to disease. With the exception of a few specific instances, as later discussed, the variety 31-1389 has in general remained resistant to diseases and may be grouped with such varieties as D 1135 and Yellow Caledonia which have been outstanding in their tolerance to sugar cane diseases.

Before discussing the reaction of 31-1389 to the various diseases it might be well to review its parentage, which follows:

$$\begin{array}{lcl}
 31-1389 \dots\dots & \left\{ \begin{array}{l} \text{POJ 2878} \\ \times \\ 26 \text{ C 270} \dots\dots \end{array} \right. & \left\{ \begin{array}{l} \text{Yel. Cal.} \\ \times \\ \text{H 109} \dots\dots \end{array} \right. \left\{ \begin{array}{l} \text{Lahaina} \\ \times \\ ? \end{array} \right.
 \end{array}$$

It is also of interest to consider 31-1389 and its parent varieties in relation to the major diseases; this information is presented in tabular form:

Major diseases	Varieties					
	31-1389	POJ 2878	26 C 270	Yel. Cal.	H 109	Lahaina
Brown stripe .....	—	—	—	—	—	—
Chlorotic streak .....	=	—		=	= +	
Eye spot .....	+ +	—	+	+	—	—
Leaf scald .....	= +	+ +		= —	=	
Mosaic .....	+ +	+ +	+	+	=	—
Pythium root rot.....	+	+	=	+	+	— —

In the above table each variety has been considered separately in its reaction to each disease and is so indicated by the following symbols :

+	+	Very highly resistant
+		Highly resistant
=	+	Moderately resistant
=		Average
=	-	Moderately susceptible
-		Highly susceptible
-	-	Very highly susceptible

Of the six varieties, Lahaina and H 109 have shown the greatest susceptibility to the major diseases while 31-1389 has shown the greatest resistance; the varieties Yellow Caledonia and POJ 2878 follow 31-1389 in their tolerance to the various diseases. It is interesting to compare the high degree of resistance of 31-1389 to specific diseases with the low degree of resistance to the same diseases of some of the other varieties entering into its parentage. The above gradings are based on field observations and disease resistance tests. Where no symbol is given in the table there is little or no available information regarding the reaction of the variety to disease.

From the first experiments, 31-1389 was found to be highly resistant to eye spot disease and its spread to those localities where the disease has repeatedly occurred has controlled eye spot and given greater protection to surrounding fields. The ultimate economic control of eye spot can only be accomplished by substituting resistant and high-sucrose varieties for the susceptible ones—the planting of 31-1389 has accomplished much toward this end.

According to our records no case of mosaic disease has been observed on 31-1389; its resistance is even better than that of POJ 2878 on which only two cases of the disease have been reported since it was introduced into the Territory in 1928.

The incidence of chlorotic streak in localities favorable for the disease has been moderate on 31-1389; field observations show it to be more resistant than POJ 2878 under similar environmental conditions.

On windward Kauai the incidence of brown stripe has been moderate to severe on 31-1389. In some instances the lesions have coalesced thus causing the older leaves to manifest a prematurely dried appearance. On Oahu the degree of brown stripe infection has not been serious while on Maui and Hawaii the disease has been of less importance.

A leaf spotting characterized by small, irregular, chlorotic markings which later assume a light brownish color is frequently noted on 31-1389. The spotting is most pronounced on the older leaves and no doubt interferes somewhat with the normal development of affected plants. Our studies to date indicate that the disease is due to some physiological cause; no organism has been associated with the spots. In nutrient solution studies this particular leaf spot failed to develop on 31-1389 when grown in solutions lacking each of the following elements: nitrogen, potassium, phosphorus, magnesium, calcium, sulphur, manganese, iron, and boron.

According to our field observations and resistance tests, 31-1389 has been placed in the group of canes resistant and tolerant to leaf scald disease. An occasional case of leaf scald disease has been observed on 31-1389. It should be mentioned, however, that an epidemic of leaf scald disease developed in one small planting of

31-1389 on Hawaii. With the exception of this one outbreak of leaf scald, the severity of the disease on 31-1389 has remained of minor importance.

The variety 31-1389 has been grown in districts where other varieties have been moderately to severely attacked by *Pythium* root rot but it has remained unaffected. A small amount of ratoon chlorosis has developed on 31-1389 but here again it has proved to be more tolerant than H 109. Ring spot at times appears on the older leaves of 31-1389 but this disease has been of little significance. No economic losses from pokkah boeng have occurred on 31-1389 but an occasional case of the disease has been noted on the variety.

Little or no losses have occurred on 31-1389 from red rot or red stripe disease. At the higher elevations, especially during the winter months, leaf freckle has appeared on the older leaves of 31-1389 but this leaf spotting has been less severe than on D 1135, POJ 36 and other varieties.

## 31-1389—Its Susceptibility to Insect Attack in Hawaii

By C. E. PEMBERTON

The variety 31-1389 has exhibited no special susceptibility to attack by any of the sugar cane insects in Hawaii. Fieldmen have noticed that it remains unusually free of cane aphid and coincident black sooty mold during seasons when other varieties in the same locality are so affected.

It is less damaged by beetle borer than the variety POJ 2878 and is at least equal to H 109 in this respect. Although the rind is softer than in H 109, the self-stripping habit and slower growth in the second season seem to give 31-1389 a better resistance to borer attack than other varieties equally soft.

## 31-1389—Its Response to Fertilizers

By R. J. BORDEN

An examination of the results of cooperative fertilizer tests which have been conducted on the variety 31-1389 indicates that this cane is probably a very economical and efficient user of plant food, especially of nitrogen.

Of the 38 harvest results which we have recorded from Grade A "Amounts-of-Nitrogen" tests on this cane variety, we note that in 30 cases no reliable gain in sugar yield was secured for amounts above the minimum application of nitrogen that was included in the test; hence in these cases it is impossible to tell just how little nitrogen would have produced the optimum sugar yields. But a study of the accompanying Table I, in which the pertinent results are summarized, will quite clearly indicate that this variety has not made efficient use of the larger applications (more than 200 pounds) of nitrogen to put "sugar in the stalk."

Two experiments comparing ammonium sulphate with nitrate of soda on 31-1389 at The Lihue Plantation Company, Ltd., and a similar test at Kekaha Sugar Company showed no differences in the effectiveness of these two nitrogen carriers.

An experiment at Pepeekeo Sugar Company, on the time to apply 175 pounds of N to this variety, indicated significantly greater cane yields (by  $8.2 \pm 1.7$  T.C.A.) when 50 pounds was applied at 7 weeks, another 50 pounds at 4 months, and 75

pounds at 6 months than when the total application was completed at 4 months. The quality was not affected by the time of application. At Waipio substation, 150 pounds of nitrogen applied at  $1\frac{1}{2}$  months was as effective as when divided into three 50-pound doses and applied at  $1\frac{1}{2}$ ,  $3\frac{1}{2}$ , and  $8\frac{1}{2}$  months.

Eleven "Amounts-of-Phosphate" experiments, all except one from the island of Kauai and installed on soils which have been considered as being deficient in available phosphoric acid, have failed to show any response to applications of phosphate fertilizer (see Table II).

From twenty "Amounts-of-Potash" experiments (summarized in Table III), most of which were located on soils which are considered to be deficient in available potash, we find only 7 cases in which there was any reliable response to potash fertilization. At Honomu Sugar Company, on a  $14\frac{1}{2}$ -month ratoon crop harvested in August, there was a response only up to 125 pounds of  $K_2O$  per acre. At The Lihue Plantation Company a 20-month plant crop harvested in December, and its subsequent  $16\frac{1}{2}$ -month ratoon cut in May, both responded to a 200-pound application—the maximum amount included in the test. However, in another test at The Lihue Plantation Company a plant crop cut at 18 months in July had shown no response, and the response on its subsequent ratoon which was cut at 19 months in February was only to 100 pounds per acre. At Grove Farm Company, a 19-month ratoon crop cut in March responded to only 100 pounds. At Kahuku Plantation Company a 16-month plant crop cut in July gave a response up to 200 pounds per acre, while its subsequent 19-month ratoon cut in February responded to only 100 pounds; in both this test and in the Grove Farm Company experiment, there was no effect from a difference in the time of applying the potash. At Waimanalo Sugar Company an indicated response to potash was not statistically verified.

If the conditions under which these fertilizer experiments have been conducted can be considered as generally representative of the areas where this variety is being grown, the results would indicate that plantation fertilizer practices which have been devised for other cane varieties may need some revision if best results are to be secured from the variety 31-1389.

TABLE I  
SUMMARY OF GRADE A "AMOUNTS-OF-NITROGEN" TESTS CONDUCTED ON VARIETY 31-1389

Plantation	Expt. No.	Crop year	Month harvested	Age (mos.)	Amounts compared (lb.)	Variables applied months	Optimum** T.S.A. secured from	From this optimum amount of nitrogen- yields secured T.C.A. T.S.A.	lb N used per acre per month	lb N used per ton sugar
Kohala Sugar Co.	108 AN	1939	Feb.	18	75-125-175	7 and 8	125 lb N	79 8.0	7.0	15.6
"	109 AN	1939	Feb.	18	100-150-200	6 and 7	100 lb N*	82 7.8	5.6	12.8
Pauhan S. Plt. Co.	65 AN	1938	Sept.	16½	75-100-150	7	75 lb N*	53 6.8	4.5	11.0
Pepeekeo Sugar Co.	40 AN	1938	Aug.	16	75-125-175-225	2, 4 and 6	75 lb N*	52 5.8	4.7	12.9
Honouu Sugar Co.	60 AN	1937	June	15	75-125-175	4 and 5	125 lb N	45 5.3	8.3	23.6
"	60 AN(R)	1938	Oct.	16	75-125-175	3, 5 and 7	125 lb N	58 6.9	7.8	18.1
H. C. & S. Co.	69 AN	1938	July	27	180-230-280	2, 5 and 6	180 lb N*	100 12.1	6.7	14.9
Kaeleku Sugar Co.	27 AN	1939	Feb.	15½	0-75-150	2, 4 and 7	75 lb N	56 4.8	4.8	15.6
"	26 AN(R)	1939	May	17	50-100-150	1, 3 and 5	150 lb N	71 5.3	8.8	28.3
Waipio Substation	21 AN	1937	June	18	100-150-200	2, 3 and 6	150 lb N	74 10.1	8.3	14.8
"	21 AN(R)	1939	June	24	100-150-200	1, 4 and 9	100 lb N*	87 11.2	4.2	8.9
Waimanalo Sugar Co.	54 AN	1937	Sept.	14	150-200-250	1, 4 and 6	150 lb N*	52 6.2	9.3	24.2
"	54 AN(R)	1939	Jan.	16½	150-200-250	2, 4 and 5	150 lb N*	84 7.0	9.1	21.4
"	57 AN	1939	May	20	200-250-300	2 and 3	200 lb N*	80 9.7	10.0	20.6
Kahuku Plt. Co.	68 AN	1936	Sept.	16	125-175-225	2 and 4	125 lb N*	90 7.7	7.8	16.2
"	68 AN(R)	1938	April	18½	125-175-225	2 and 7	125 lb N*	87 8.9	6.8	14.0
"	86 AN	1939	July	21½	106-169	6	106 lb N*	48 5.8	4.9	18.3
Honolulu Plt. Co.	63 AN	1938	April	19	150-250-350	2, 3, 4 and 6	150 lb N*	109 11.2	7.9	13.4
"	66 AN	1939	Aug.	17	150-250-350	2, 4 and 6	150 lb N*	85 8.6	8.8	17.5
Kilauea S. Plt. Co.	205 AN	1938	March	20½	150-250-350	2, 4, 6 and 7	150 lb N*	104 12.6	7.3	11.9
"	205 AN(R)	1939	June	15	125-175-225-275	2, 3, 4 and 6	125 lb N*	81 6.5	6.3	19.3
Grove Farm Plt. Co.	70 AN(R)	1939	April	19	100-200	1, 2, 3 and 5	125 lb N*	55 3.9	8.5	32.1
"	140 AN	1936	Dec.	20	150-200-250	2 and 5	100 lb N*	76 9.4	5.3	10.6
"	140 AN(R)	1938	May	16½	150-200-250	1 and 2	200 lb N	90 8.6	10.0	23.2
"	152 AN	1937	June	22½	128-178-228	3 and 5	150 lb N*	66 8.2	9.1	18.3
"	152 AN(R)	1939	March	20½	125-175-225	1 and 2	128 lb N*	76 10.3	5.7	12.4
"	155 AN	1937	July	18½	150-200-250	3	125 lb N*	79 9.5	6.1	13.2
"	155 AN(R)	1939	Feb.	19	163-213-263	3 and 6	150 lb N*	75 7.4	8.1	20.2
"	166 AN	1937	Aug.	17	150-200-250	3 and 4	163 lb N*	77 8.8	8.6	18.5
"	166 AN(R)	1939	Feb.	18	142-192-242	5 and 6	150 lb N*	73 6.7	8.8	22.4
"	183 AN	1938	June	17	150-200-250	2	142 lb N*	80 9.8	7.9	14.5
Kekaha Sugar Co.	14 AN	1937	Oct.	17	160-200-240-280	3 and 4	150 lb N*	73 7.5	8.8	20.0
"	14 AN(R)	1939	March	17	150-200-250	4, 6 and 8	160 lb N*	83 8.5	9.4	18.8
Pioneer Mill Co.	328 AN	1937	May	21½	100-200-250	5, 7, 8 and 10	150 lb N*	84 9.0	8.8	17.0
"	322 AN	1937	Dec.	20	150-200-250-300	2, 4, 5 and 7	100 lb N*	88 8.9	4.8	11.2
"	324 AN	1937	May	23½	150-200-250-300	3, 10 and 11	200 lb N	112 12.3	10.0	16.2
"	336 AN	1937	Dec.	17	156-208-250	3, 5, 8 and 10	150 lb N*	94 10.2	6.4	14.7
"	336 AN	1937	Dec.	17	156-208-250	8 and 9	156 lb N*	77 6.1	9.2	25.6

\* The minimum amount used in the experiment.

(R) Ratoon crop.

\*\* No significant gain in T.S.A. secured for greater amounts of N per acre.

TABLE II  
SUMMARY OF GRADE A "AMOUNTS OF PHOSPHATE" TESTS CONDUCTED ON VARIETY 31-1389

Plantation	Expt. No.	Crop year	Month harvested	Age (mos.)	Amounts compared (lb)	Variables applied at months	Optimum* T.S.A. secured from	From this optimum amount of phosphate yields secured T.C.A.
Pioneer Mill Co.....	326	1937	May	22½	0-100-200-300	1	No P <sub>2</sub> O <sub>5</sub>	88 10.3
Grove Farm Co.....	65 (R)	1939	March	19	0-200	1 and 6	" "	76 8.5
Kilauea S. Plt. Co.....	203 (R)	1939	June	15	0-100-200-300	1	" "	55 3.8
Lihue Plt. Co.....	141	1936	Dec.	20	0-125-250	At planting	" "	96 8.6
" " " " " "	141 (R)	1938	May	16½	0-100-200	3	" "	64 8.0
" " " " " "	154 (R)	1939	March	20	0-125-250	1	" "	77 9.8
" " " " " "	157 (R)	1939	Feb.	19	0-125-250	1½	" "	78 9.2
" " " " " "	184	1938	June	17	0-100-200	2	" "	69 7.2
" " " " " "	195	1939	Feb.	19½	0-100-200	1¾	" "	79 9.7
McBryde Sugar Co....	42	1939	April	18	0-200	1¾	" "	113 9.8
Waimanalo Sugar Co....	58	1939	June	14½	0-180	At planting	" "	61 5.2

\* No significant gain in T.S.A. secured for greater amounts of P<sub>2</sub>O<sub>5</sub> per acre.

(R) Ratoon crop.

TABLE III  
SUMMARY OF GRADE A "AMOUNTS-OF-POTASH" TESTS CONDUCTED ON VARIETY 31-1389

Plantation	Expt. No.	Crop year	Month harvested	Age (mos.)	Amounts compared (lb)	Variables applied at months	Optimum* T.S.A. secured from	From this optimum amount of potash yields secured T.C.A.
Honoum Sugar Co.....	59 (R)	1938	Aug.	14½	0-125-250-375	3, 5 and 7	125 lb K <sub>2</sub> O	56 5.3
Pepeseko Sugar Co.....	41	1938	Aug.	16	0-100-200-300	2, 4 and 6	No K <sub>2</sub> O	58 5.7
Kaeleku Sugar Co.....	28 (R)	1939	March	15	0-150-300	1, 3 and 6	" "	43 4.4
Pioneer Mill Co.....	325	1937	May	22½	0-100-200-300	1	" "	95 10.4
" "	345	1938	Feb.	18½	43-101	7	43 lb K <sub>2</sub> O	102 8.1
Kahuku Plt. Co.....	80	1937	July	16	0-100-200	1, 4	200 lb K <sub>2</sub> O	44 4.1
" "	80 (R)	1939	Feb.	19	0-100-200	2	100 lb K <sub>2</sub> O	45 5.1
Grove Farm Co.....	66 (R)	1939	March	19	0-100-200	1	100 lb K <sub>2</sub> O	73 8.4
Kilauea S. Plt. Co.....	204	1938	March	20	0-125-250	1 and 5	No K <sub>2</sub> O	65 5.5
" "	204 (R)	1939	June	15	0-125-250	1	" "	55 3.5
Lihue Plt. Co.....	142	1936	Dec.	20	0-100-200	1	200 lb K <sub>2</sub> O	102 9.3
" "	142 (R)	1938	May	16½	0-100-200	3	200 lb K <sub>2</sub> O	66 8.4
" "	153	1937	July	23	0-100-200	1	No K <sub>2</sub> O	80 11.3
" "	153 (R)	1939	March	20	0-100-200	1	" "	78 9.7
" "	156	1937	July	18	0-100-200	At planting	" "	70 6.6
" "	156 (R)	1939	Feb.	19	0-100-200	1½	100 lb K <sub>2</sub> O	79 9.5
" "	185	1938	June	17	0-100-200	2 and 7	No K <sub>2</sub> O	74 7.6
" "	196	1939	Feb.	19½	0-100-200	1½	" "	80 10.0
McBryde Sugar Co.....	43	1939	April	18	0-100-200	2½	" "	112 9.3
Waimanalo S. Co.....	58	1939	June	14½	0-213	2 and 5	" "	61 5.2

\* No significant gain in T.S.A. secured for greater amounts of K<sub>2</sub>O per acre.  
(R) Ratoon crop.

## 31-1389—Its Manufacturing Qualities

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By W. L. McCLEERY

The year 1939 is the third year that questionnaires have been sent to the plantations asking for specific information on the manufacturing qualities of new cane varieties that are being harvested in appreciable quantities. Below is a summary of replies received to September 1, 1939 on variety 31-1389. The figures for acres to be harvested in 1939 are taken from the Genetics Department, "An Acreage Census of Cane Varieties for the Crops of 1938 and 1939," dated March 1939. The percentages of crops harvested are from the 1938 and 1937 "Annual Synopsis of Mill Data."

*Hakalau:*

1939: Acres to be harvested, 63. No report received to the date of this compilation.

*Hilo:*

1939: Acres to be harvested, 119. All factory qualities good, compared with the standard varieties POJ 36 and Yellow Caledonia; also less trash.

*Honolulu:*

1939: Acres for harvest, 1452. Cane ground mixed with other canes. No outstanding poor qualities.

1938: Was 43 per cent of crop. No poor qualities.

1937: Was 19 per cent of harvest. Reported good, except for bagasse steaming quality which was fair.

*Honoumū:*

1939: Acres for harvest, 640. No report to this date.

*Hutchinson:*

1939: Acres for harvest, 37. No report.

*Kaeleku:*

1939: Acres for harvest, 453. Rated as fair in boiling house and poor in mill-feeding and bagasse-burning qualities, as compared with POJ 2878. Has less trash; also lower fiber.

1938: Was 8 per cent of harvest. Ground mixed.

1937: Was 10 per cent of harvest. Rated as fair; poor in mill-feeding.

*Kahuku:*

1939: Acres for harvest, 455. Rated good throughout. After grinding considerable quantities there appears to be no appreciable difference at the mill or boiling house as compared with H 109.

1937: Rated as fair on first runs from poorly drained land.



*Kaiwiki:*

1939: Acres for harvest, 55. Has slightly more trash than D 1135. Good milling quality and is fair as a steam producer and in the boiling house. Given general rating of good.

*Kekaha:*

1939: Acres for harvest, 215. No report.

1937: Rated as fair to poor throughout.

*Kilauea:*

1939: Acres for harvest, 417. Several 100 per cent runs expected. Report not received.

1938: Ground 14 per cent of this cane for year. While it was generally mixed with POJ 2878, there was no trouble encountered. Rated as good throughout.

1937: Was 5 per cent of harvest. Rated as good. Better than either POJ 2878 or Badila.

*Kohala:*

1939: Acres this year's harvest, 801. Ground mixed; no definite information. No opinions.

1938: Rated to be just as good as other varieties. Was 3 per cent of crop.

*Lihuc:*

1939: Acres, including Grove Farm, 1523. No report.

1938: Was 10 per cent of crop. Was good at the mill but poor in steam quality. No opinions for boiling house as it was ground mixed with other canes.

1937: Was 7 per cent of crop. No opinions possible as it was mixed with too much field soil.

*Maui Agric.:*

1939: Acres for harvest, 216. No report.

*McBryde:*

1939: Acres for 1939 harvest, 248. Rated as fair throughout, but conditions were not normal for accurate judging.

1938 and 1937: Was 4 per cent of each crop. Rated fair-to-good, each year.

*Oahu:*

1939: Acres for 1939 season, 442. No report received.

1938: Was 3 per cent of crop. Ground mixed; no opinions.

1937: Was 1 per cent of crop. Preliminary indications were that it had fair mill-feeding and poor steaming qualities.

*Paaunhau:*

1939: Acres for harvest, 153. Rated as good. Grinding rate is increased due to large stick and less trash. The fiber is lower and necessitates faster grinding to maintain steam and boiler draft adjustments. It has a high phosphate content which makes ample settling capacity needed, especially when cane is mechanically harvested.

*Pepeekeo:*

1939: Acres for harvest, 277. No report received.

*Pioneer:*

1939: Acres for harvest, 518. No report received.

1938 and 1937: Constituted 8 to 9 per cent of each year's harvest. Rated good to fair in most qualities, but poor as steam producer.

*Waialua:*

1939: Acres for harvest, 466. No report.

1938 and 1937: Constituted 6 and 4 per cent respectively of tonnage ground each year. Ground mixed so opinions preliminary. Indications were poor qualities throughout, especially as steam producer.

*Waianae:*

Very little of this cane for 1939 and in 1938. In 1937 a large field gave very poor qualities throughout the factory. This field was replanted with other cane.

*Wailuku:*

1939: Acres for harvest, 834. No report yet available.

1938: Was 20 per cent of harvest. No general opinion; had fair mill-feeding and poor steaming quality.

1937: Was 4 per cent of crop. Ground mixed with POJ 2878 in rainy weather and no differences were noted.

*Waimanalo:*

1939: Acres for harvest, 1161. Has replaced POJ 2878 as standard variety. Good factory qualities throughout. As with POJ 2878, to obtain sufficient steam, draft and furnace adjustments were necessary.

1937: Was 12 per cent of crop. Rated as satisfactory.

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## A Lysimeter Study of Losses of Applied Potash by Leaching From an Acid Soil

By P. L. Gow

### INTRODUCTION

This experiment was designed as part of a project which has as its purpose the evaluation of probable injurious effects upon Hawaiian cane lands of long continued fertilization with acid-forming fertilizers such as sulfate of ammonia.

Other investigators working with greatly differing soil types, and crops in widely separated parts of the world have found marked deleterious effects resulting with soils receiving continued amendments of ammoniacal salts as fertilizers. No attempt is made herewith to present a review of the literature concerning this problem since an excellent review, together with an extensive bibliography has been presented by Morgan (7, p. 381) who summarized the results of previous investigators as follows:

That fertilizer constituents supplying nitrogen as ammonium salts have an acid effect upon the soil is clearly established. In all cases where soil changes resulting from the use of such materials have been studied, one or more of the following phenomena were exhibited:

- a. Increase in hydrogen-ion concentration of the soil (decrease in pH).
- b. Increase in exchangeable hydrogen.
- c. Decrease in the exchangeable bases, especially calcium.
- d. Increase in easily soluble aluminum, iron or manganese.

Except on soils containing large reserves of calcium carbonate, such as the heavily chalked soil at Rothamsted, England, these results are readily measured within a short time, when substantial annual applications are made.

Eckart (3), working with Makiki soil in lysimeters planted to cane, found that greater quantities of calcium were leached from sulfate of ammonia treatments than were leached where nitrate of soda was applied. Peck (8) conducted lysimeter experiments with an acid upland soil from Hawaii and a basic lowland soil from Kauai. The lysimeters were not cropped. More potash and calcium were leached from the acid soil when sulfate of ammonia was applied than where no fertilizer was used. More calcium, but not significantly more potash, was leached from the basic soil following application of ammonium sulfate.

In considering the results obtained by workers elsewhere and by Eckart and Peck with Hawaiian soils it may be unwise to assume without further studies that such effects are certain to occur in soils which are cropped to sugar cane. Hawaiian cane lands support perhaps the largest masses of vegetative growth that have ever been produced on cultivated fields, and these consist of a crop which appears to have extensive effects upon the soils supporting it. Unpublished soil pH studies made by the writer at this Experiment Station in connection with Project C-3 in 1930-31 indicate that sugar cane may act as a buffer to soil pH changes, and that where fertilizers have been applied which produce either increase or decrease of soil pH, the crop, as it develops, tends to restore the pH to a value approaching that of untreated soil. This proved especially true in cases where

the soil pH was depressed as a result of fertilization with mixtures containing ammonium salts. Perhaps further investigations will indicate that effects heretofore observed elsewhere with acid-forming fertilizers may not be so significant where cane crops are grown.

However, for the purposes of this investigation, it was tentatively assumed that all of the effects mentioned by Morgan (7) would occur in soils cropped to cane. The question then became: How serious will these effects be with respect to the continued ability of Hawaiian soils to support cane crops?

Theoretical applications of soil chemistry, together with certain practical knowledge of cane requirements, lead to the following considerations:

(a) Decrease in soil pH *per se* will probably exhibit no deleterious effect upon cane yields until a very low level is reached. It can not be predicted how low that level will be without further study, since workers with Hawaiian soils have not yet found an agricultural soil of sufficiently low pH to impair seriously cane yields. Ayres (2) obtained normal cane growth in pots with soil of pH as low as 4.6, which is considerably lower than that of any Hawaiian soils normally cropped to cane. Martin (6) found that cane made good growth in water cultures at pH 4.0, but that growth was greatly retarded at pH 3.0. It is doubtful that any Hawaiian soil could attain a pH below 4.0.

(b) Increase of exchangeable hydrogen means decrease in effective exchange capacity, since hydrogen ion is very tenaciously retained by soils against replacement by bases. Hence, the capacity of soils to take up and retain bases applied as fertilizer amendments against loss by leaching might be seriously impaired as the result of ammoniacal fertilization. This effect would be especially harmful in the case of potash which is applied extensively in cane cultivation and might result in considerable economic losses.

(c) Decrease in exchangeable bases might be serious, especially in the case of potash, provided the potash which was released were leached instead of being taken up by the crop. Decrease in exchange calcium might be undesirable if it resulted in lowering the calcium reserve to a level insufficient for crop needs, or if sufficient calcium were made available to impair the juice quality of the crop. On the other hand, it is probable that sufficient calcium could be supplied as fertilizer amendment to provide for proper calcium nutrition at quite reasonable cost. In some cases, decrease of exchange calcium to a level requiring small lime applications might result in better juice quality since the calcium supply could then be controlled. Magnesium could also be supplied, presumably at low cost, as fertilizer. It is worthy of note that forms-of-nitrogen experiments have not hitherto showed poorer juice quality due to excess calcium liberated by sulfate of ammonia.

(d) Increase in easily soluble aluminum might result in toxic effects, but such effects have not hitherto been felt in forms-of-nitrogen experiments. Increase in easily soluble iron and manganese would perhaps be desirable. Should iron and manganese supplies become depleted, small applications as fertilizer amendments would be possible.

All of these effects appear possibly deleterious to Hawaiian soils, but none appear dangerous at present, except, perhaps, a decrease in effective exchange

capacity and resultant inability of soils to retain applied potash against loss by leaching. It was therefore decided to study first potash losses by leaching, together with the possibility of obviating them by the use of a basic potash salt to promote absorption by that portion of the exchange complex which was occupied by hydrogen ion. The present experiment was designed with that object in view.

Potassium carbonate was selected as the basic salt of potash to be used. While the cost per unit of potash as carbonate is about three times that of the sulfate, there is reason to believe that sources of cheaper carbonate might be developed provided the value of its use as fertilizer could be demonstrated. Preliminary laboratory tests comparing potassium carbonate with potassium chloride indicated that the former salt caused very much greater uptake of potassium ion with acid soils, due to neutralization of the exchangeable hydrogen ion by the carbonate ion and removal by leaching as  $\text{H}_2\text{CO}_3$  and by decomposition of the latter into  $\text{CO}_2$  and water. In this way the soil acidity was utilized to promote the desired effect which it tended to prevent when neutral potash salts were applied.

Muriate of potash was selected as the neutral salt to be compared, since the presence of considerable quantities of sulfate ions was considered undesirable in view of the method which was proposed to be used for analysis of the leachates.

#### EXPERIMENTAL PROCEDURE

A free-draining, acid Manoa soil of pH 5.3 was filled into cubical concrete lysimeters whose dimensions were 2 x 2 x 2 feet. The pots were heavily tarred with roofing asphalt to prevent leakage and were provided with a single drainage hole at the bottom. The bottom sloped from all sides to the drainage hole. Leachates were collected in specially constructed 5-gallon tins which were fitted with 1.5-inch openings to minimize evaporation.

The lysimeters were arranged in 4 rows of 9 pots each and were sunk to ground level. Between each set of 2 rows was a sunken concrete runway which served to allow collection of leachates. A layout of the experimental arrangement is given below:

#### LAYOUT OF EXPERIMENT

1	2	3	4	5	6	7	8	9
5	6	2	8	3	7	4	9	1
C	NV	C	NV	C	NV	C	NV	C
10	11	12	13	14	15	16	17	18
1	7	9	4	8	5	6	2	3
C	NV	NV	C	NV	C	NV	C	C
19	20	21	22	23	24	25	26	27
3	8	5	6	1	7	2	9	4
C	NV	C	NV	C	NV	C	NV	C
28	29	30	31	32	33	34	35	36
6	4	7	2	9	3	8	1	5
NV	C	NV	C	NV	C	NV	C	C

The upper numbers which run consecutively are pot numbers. The lower numbers are treatment numbers. The designations C and NV indicate whether the pot was planted to cane or was kept free of vegetation.

### *Fertilizer Treatments:*

In addition to comparing the leaching losses of potassium carbonate and chloride, it was thought desirable to compare the effects of applying potash together with ammoniacal salts and of delaying application of potash until nitrification of the ammonium salts was completed. It appeared likely that, even though potash from the carbonate was forced into the base-exchange complex due to neutralization of exchange hydrogen, subsequent production of hydrogen ions by nitrification of ammonium salts might result in release of potash initially absorbed. Somewhat greater loss of potash from the muriate of potash treatments might also be expected from this cause. Experience of other workers had demonstrated that 45 days was ample to secure complete nitrification of ammonium salts in Manoa soils of the same type and originating from the same locality as the soil used in this experiment. Therefore it was decided to include carbonate and muriate of potash treatments, in which potash application was withheld for 45 days after nitrogen was applied. Ammonium chloride instead of sulfate was used as nitrogen application for the same reason that the chloride of potassium was selected.

Treatments in which pots were kept free of vegetation were included, since it was considered essential, in view of previous experience, to determine whether the presence of a cane crop materially influenced the results.

In view of the fact that by far the greater proportion of Hawaiian cane crops are ratoon rather than plant crops, this experiment was conducted with ratoon cane. To secure uniformity of stand, four 3-eye cuttings of H 109 cane were planted in each of the 36 pots. After the shoots were well started, three of the cuttings were dug up and rejected, while two shoots from the third cutting were excised. Thus a quite uniform stand of cane was started, each pot containing one shoot attached to a 3-eye seedpiece. After the cane had been allowed to grow for four months with daily irrigation, the cane was cut at ground level in the pots to be ratooned and was cut back to the seedpiece in the no-vegetation pots. At this time enough stalk had developed in each pot to insure a vigorous ratoon crop. On July 5, 1938, fourteen days after harvest, the first application of fertilizer was made and collection of the first series of leachate samples begun. At this time all pots received nitrogen at the rate of 100 pounds N per acre as  $\text{NH}_4\text{Cl}$ . Previously, when the soil was filled into the pots, all pots had received 100 pounds N per acre and 369 pounds  $\text{P}_2\text{O}_5$  as Ammo-phos 13-48.

Fertilizer applications were made in solution and were applied to an area one foot long and three inches wide on a line directly above the seedpiece. In the field, it is estimated that there normally occurs about one stool of cane per foot of line; counts made in experimental plots have indicated that about four fully grown stalks of cane per foot represent a heavy growth of cane. Cane growth was therefore limited to four stalks per pot. According to field practice in Hawaii, cane lines are spaced 5 feet apart, so each stool of cane averages about 5 square feet of surface. In this experiment each stool enjoyed only 4 square feet of surface, due to the size of the lysimeters. Each pot was considered as representing one running foot of line and a figure of 8712 running feet of line per acre was accepted as basis for calculation of fertilizer applications.

On July 5, at the same time that all treatments received nitrogen as ammonium chloride, the "potash-with-nitrogen" treatments received 300 pounds per acre of  $K_2O$  as either muriate or carbonate. Forty-five days later the "potash-after-nitrogen" treatments received a like amount of potash.

The various treatments were as follows:

#### CANE TREATMENTS

1. No  $K_2O$ .
2. KCl with nitrogen.
3.  $K_2CO_3$  with nitrogen.
4. KCl after nitrogen.
5.  $K_2CO_3$  after nitrogen.

#### NO-VEGETATION TREATMENTS

6. KCl with nitrogen.
7.  $K_2CO_3$  with nitrogen.
8. KCl after nitrogen.
9.  $K_2CO_3$  after nitrogen.

#### *Irrigation:*

In order to compare the cane treatments with the no-vegetation treatments, it was decided, instead of irrigating all pots alike, to apply water in such manner as to cause all pots to yield, as nearly as possible, the same amounts of leachate. This, of course, required an application of greater quantities of water to the cane pots than to the no-vegetation pots in order to compensate for the water consumed by the cane. The technic of irrigating to this end was developed during the time the plant crop was being grown to the point at which it could be safely ratooned. It was sought to make each pot approximate an average leaching of 3 liters per day which would be equivalent to 0.32 inch. The water consumption for each day of each pot, together with rainfall data, was used to calculate the required irrigation for the following day. Unfortunately, at the end of each 45-day sampling period, several days of rainy weather occurred which made it impossible to secure strictly comparable amounts of leachates for the two periods. The amounts of leachates from the various pots at the end of each period are fairly comparable (Table I). Irrigation was with tap water and each day a sample representing 3 liters was taken. The amounts of  $K_2O$  in the tap water given in Table I represent the summation of the amounts of  $K_2O$  in successive 3-liter per day portions taken over each 45-day period. The daily irrigations for each pot were weighed out by means of a solution balance.

#### *Sampling:*

The water leached from each lysimeter was collected in a 5-gallon tin can which was especially constructed to minimize evaporation and to exclude rainwater. Samples were usually collected every other day. However, in rainy weather it was necessary to sample daily and a few times, when conditions were favorable,



as much as 4 days' leachates were allowed to collect before sampling. The procedure followed in sampling was to weigh each can with the contained leachate, shake the can vigorously to insure uniformity of sample, take one-tenth of the leachate for analysis, empty the can and weigh the can again for tare.

#### *Analysis of Leachates:*

The leachate samples, representing one-tenth of the total leachate for each 45-day period, were evaporated on a hot plate and aliquots were taken for determination of  $K_2O$ . Potash was determined colorimetrically by the Hill method (5) as modified by Gow (4).

#### DISCUSSION OF DATA

Table I shows the amount of  $K_2O$  leached from each pot during each of the 45-day periods, together with the total amount for the entire 90 days. The leachate sample from Pot 18, Series A, was lost so that the value given here is an estimated value calculated statistically by the method of Allan and Wishart (1).

Table II gives the mean for each series and for the total amounts of potash leached. Appended to Table II is a table showing the differences required for significance for each series. Since it was desired to compare the averages of Treatments 2 and 3 in Series A with 4 and 5 in Series B, and 6 and 7 in Series A with 8 and 9 in Series B, the required differences for these comparisons are also given in the column labeled "A and B."

Table III is presented to show that there were no significant differences due to position of pots between the various treatments.

In Table IV are given the means for the various treatments of the per cent of the applied  $K_2O$  which was leached. These values are calculated by subtracting mean amounts leached from the No- $K_2O$  treatments in each series from the means for each of the other treatments, dividing by the amount of potash applied and multiplying by 100.

Table V-A lists the differences between the treatments within each series to facilitate comparison of the various average amounts leached.

In order to discover the effect of delaying application of potash until nitrification of ammonium salts is complete it is necessary to compare Treatments 1A, 2A, 3A, 4B, 5B, 6A, 7A, 8B and 9B since these represent the 45-day periods immediately following potash application. Table V-B gives the differences between these averages, which are not shown by Table V-A.

#### *Cane Treatments:*

Treatment 2 showed no significantly increased leaching over Treatment 1 during the first 45 days following application of potassium chloride together with ammonium sulfate. During the second 45-day period, however, an average of 0.88 gram more  $K_2O$  was leached from these pots than from the no-potash pots. A difference of 0.75 gram is required for odds of 19 to 1, so this increase is significant although not highly so. The mean loss from Treatment 2 for this period represented 5.7 per cent of the potash applied.

TABLE I

Soil pH = 5.3									
Treatment	Pot No.	Inches water leached 1st set	Inches water leached 2nd set	Total inches water leached	Gms K <sub>2</sub> O applied 1st set	Gms K <sub>2</sub> O applied 2nd set	A Gms K <sub>2</sub> O leached 1st period	B Gms K <sub>2</sub> O leached 2nd period	Total Gms K <sub>2</sub> O leached
CANE									
1—No K	{ 9	16.51	14.15	30.66	0	0	.198	.560	.758
	{ 10	16.11	13.98	30.09	0	0	.648	.508	1.16
	{ 23	16.34	14.03	30.37	0	0	.436	.234	.670
	{ 35	16.95	13.96	30.91	0	0	.468	.552	1.02
2—KCl with N	{ 3	16.33	14.05	30.38	15.621	0	.198	.918	1.12
	{ 17	17.68	13.79	31.47	15.621	0	.420	1.41	1.83
	{ 25	16.59	14.08	30.67	15.621	0	.530	1.59	2.12
	{ 31	17.62	14.49	32.11	15.621	0	.766	1.45	2.22
3—K <sub>2</sub> CO <sub>3</sub> with N	{ 5	16.80	14.17	30.97	15.621	0	.234	.560	.794
	{ 18	17.01	14.17	31.18	15.621	0	.790	1.06	1.85
	{ 19	16.89	14.12	31.01	15.621	0	1.15	.350	1.50
	{ 33	16.52	14.10	30.62	15.621	0	.358	.666	1.22
4—KCl after N	{ 7	16.66	14.10	30.76	0	15.621	.856	.650	1.51
	{ 13	17.43	14.08	31.51	0	15.621	.582	.338	.920
	{ 27	16.65	14.13	30.78	0	15.621	.380	.472	.852
	{ 29	16.70	14.12	30.82	0	15.621	.464	.596	1.06
5—K <sub>2</sub> CO <sub>3</sub> after N	{ 1	16.73	14.10	30.83	0	15.621	.558	.540	1.10
	{ 15	17.04	14.29	31.33	0	15.621	.348	.258	.606
	{ 21	16.98	13.95	30.93	0	15.621	.400	.332	.732
	{ 36	17.61	14.06	31.67	0	15.621	.624	.412	1.04
NO VEGETATION									
6—KCl with N	{ 2	16.76	14.36	31.12	15.621	0	3.26	3.12	6.38
	{ 16	16.89	14.67	31.56	15.621	0	.958	3.44	4.40
	{ 22	16.79	14.35	31.14	15.621	0	2.36	3.26	5.62
	{ 28	16.54	14.31	30.85	15.621	0	2.66	5.16	7.82
7—K <sub>2</sub> CO <sub>3</sub> with N	{ 6	16.75	14.35	31.10	15.621	0	1.21	4.32	5.53
	{ 11	16.71	14.35	31.06	15.621	0	4.18	3.58	7.76
	{ 24	16.94	14.36	31.30	15.621	0	2.04	2.48	4.52
	{ 30	17.39	14.36	31.75	15.621	0	.524	2.22	2.74
8—KCl after N	{ 4	17.24	14.35	31.59	0	15.621	.852	1.48	2.33
	{ 14	16.75	14.33	31.08	0	15.621	1.12	1.57	2.69
	{ 20	17.10	14.37	31.47	0	15.621	.456	1.76	2.22
	{ 34	16.45	14.34	30.79	0	15.621	1.28	1.39	2.67
9—K <sub>2</sub> CO <sub>3</sub> after N	{ 8	16.65	14.33	30.98	0	15.621	.310	.618	.928
	{ 12	16.72	14.32	31.04	0	15.621	.390	.824	1.21
	{ 26	16.67	14.33	31.00	0	15.621	.738	1.01	1.75
	{ 32	17.08	14.36	31.44	0	15.621	.952	1.39	2.34
H <sub>2</sub> O							*.165	*.567	*.732
Avg. 16.76				Avg. 14.22	Avg. 30.98				
* These figures represent the grams K <sub>2</sub> O originally present in the average amount of irrigation water which leached through the pots. They were obtained by calculating the potash content of the water samples taken for each period to the basis of inches water leached.									

\* These figures represent the grams K<sub>2</sub>O originally present in the average amount of irrigation water which leached through the pots.

They were obtained by calculating the potash content of the water samples taken for each period to the basis of inches water leached.

TABLE II  
AVERAGE GRAMS  $K_2O$  LEACHED PER POT FROM EACH TREATMENT

Treatment No.	Treatment	Sample series		
		A	B	Total
CANE				
1	No K <sub>2</sub> O .....	.44	.46	.90
2	KCl with N.....	.48	1.34	1.82
3	K <sub>2</sub> CO <sub>3</sub> with N.....	.68	.66	1.34
4	KCl after N.....	.57	.52	1.09
5	K <sub>2</sub> CO <sub>3</sub> after N.....	.48	.39	.87
NO VEGETATION				
6	KCl with N.....	2.31	3.75	6.06
7	K <sub>2</sub> CO <sub>3</sub> with N.....	1.99	3.15	5.14
8	KCl after N.....	.93	1.55	2.48
9	K <sub>2</sub> CO <sub>3</sub> after N.....	.60	.96	1.56

Difference needed for significance	Series			
	A	B	A & B*	Total
19 to 1.....	1.02	.75	1.01	1.34
99 to 1.....	1.39	1.02	1.37	1.82

\* By Series A & B is meant a series made up of Treatments 1A (or 1B), 2A, 3A, 4B, 5B, 6A, 7A, 8B and 9B.

TABLE III  
ANALYSIS OF DIFFERENCES BETWEEN TREATMENTS DUE TO LOCATION OF POTS

Series	"F" value	
	Found	Required for significance
A .....	.12	3.01
B .....	.40	3.01
A & B.....	.17	3.01
Total .....	.18	3.01

TABLE IV  
AVERAGE PER CENT  $K_2O$  APPLIED WHICH WAS LOST THROUGH LEACHING

Treatment No.		Sample series		
		A	B	Total
CANE				
1	No $K_2O$ .....	0	0	0
2	KCl with N.....	0.3	5.7	6.0
3	$K_2CO_3$ with N.....	1.6	1.3	2.9
4	KCl after N.....	0.9	0.4	1.3
5	$K_2CO_3$ after N.....	0.3	—0.4	—0.1
NO VEGETATION				
6	KCl with N.....	12.0	21.1	33.1
7	$K_2CO_3$ with N.....	9.9	17.3	27.2
8	KCl after N.....	3.2	7.0	10.2
9	$K_2CO_3$ after N.....	1.0	3.2	4.2

Per cent  $K_2O$  applied which was leached =

$$\frac{\text{Grams } K_2O \text{ leached} - \text{Grams } K_2O \text{ leached from No-}K_2O \text{ pots}}{\text{Grams } K_2O \text{ applied}} (100)$$

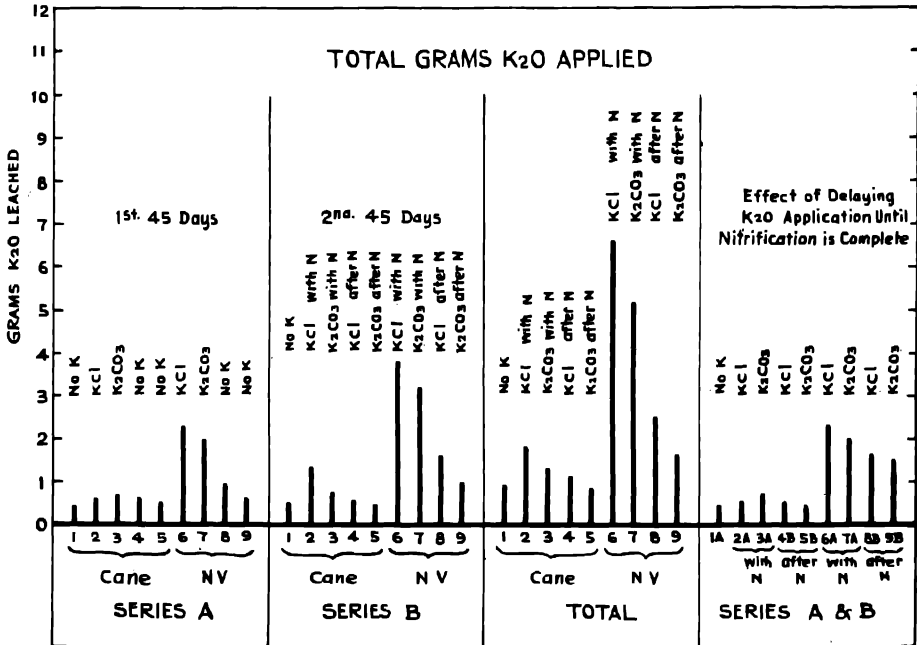


Fig. 1. Graphical comparison of amounts of potash leached from the various treatments.

The difference between the total amount of potash leached over the entire 90 days from Treatment 2 and that from Treatment 1 is 0.92 gram, which is considerably less than the amount required (1.34 grams) and is therefore not a reliable difference. However, the fact that the loss of potash from Treatment-2 pots during the second 45 days was significant indicates that more trust can be put in the total loss of 6.0 per cent of applied potash than is indicated by the statistical analysis of the totals alone.

Differences between Treatments 3 and 2 are not proved to be significant. However, neither are differences between Treatments 3 and 1, but between 2B and 1B they are reliable. In view of these facts and in consideration of the amounts leached from each treatment, the carbonate does not appear to offer much greater conservation of potash than does the chloride when applied to soil cropped to cane, although the data suggest that further experiments would continue to show somewhat smaller losses from potassium carbonate.

We are not justified in comparing Treatments 2 and 3 with 4 and 5 in the B series, since, when this set of samples was started, Treatments 2 and 3 had already had potash for 45 days, while Treatments 4 and 5 had just received potash. Had sampling from Treatments 4 and 5 been continued for an additional 45 days, further losses of potash due to treatment might have occurred as was the case during the second period with Treatment 2. No significant differences are found between 4B and 2A, or 5B and 3A, so delaying application of potash until after nitrification was complete did not appear to result in lower potash losses.

In general, it can be said that potash losses were so small as to be unimportant when potash was applied to pots containing a ratoon cane crop and that the effects of varying treatment were negligible.

TABLE V-A  
DIFFERENCES BETWEEN TREATMENTS IN GRAMS

Treatment	Sample series	Treatments								Difference required 19:1 99:1		
		1	2	3	4	5	6	7	8		9	
CANE												
1—No K <sub>2</sub> O	{ A B Total	..... ..... .....	.04 .88 .92	.24 .20 .44	.13 .06 .19	.04 .07 .03	1.87 3.29 5.16	1.55 2.69 4.24	.49 1.09 1.58	.16 .50 .66	1.02 .75 1.34	1.39 1.02 1.82
2—KCl with N	{ A B Total	.04 .88 .92	..... ..... .....	.20 .68 .48	.09 .82 .73	0.00 .95 .95	1.83 2.41 4.24	1.51 1.81 3.32	.45 .21 .66	.12 .38 .26	1.02 .75 1.34	1.39 1.02 1.82
3—K <sub>2</sub> CO <sub>3</sub> with N	{ A B Total	.24 .20 .44	.20 .68 .48	..... ..... .....	.11 .14 .25	.20 .27 .47	1.63 3.09 4.72	1.31 2.49 3.80	.25 .89 1.14	.08 .30 .22	1.02 .75 1.34	1.39 1.02 1.82
4—KCl after N	{ A B Total	.13 .06 .19	.09 .82 .73	.11 .14 .25	..... ..... .....	.09 .13 .22	1.74 3.23 4.97	1.42 2.63 4.05	.36 1.03 1.39	.03 .44 .47	1.02 .75 1.34	1.39 1.02 1.82
5—K <sub>2</sub> CO <sub>3</sub> after N	{ A B Total	.04 .07 .03	0.00 .95 .95	.20 .27 .47	.09 .13 .22	..... ..... .....	1.83 3.36 5.19	1.51 2.76 4.27	.45 1.16 1.61	.12 .57 .69	1.02 .75 1.34	1.39 1.02 1.82
NO VEGETATION												
6—KCl with N	{ A B Total	1.87 3.29 5.16	1.83 2.41 4.24	1.63 3.09 4.72	1.74 3.23 4.97	1.83 3.36 5.19	..... ..... .....	.32 .60 .92	1.38 2.20 3.58	1.71 2.79 4.50	1.02 .75 1.34	1.39 1.02 1.82
7—K <sub>2</sub> CO <sub>3</sub> with N	{ A B Total	1.55 2.69 4.24	1.51 1.81 3.32	1.31 2.49 3.80	1.42 2.63 4.05	1.51 2.76 4.27	.32 .60 .92	..... ..... .....	1.06 1.60 2.66	1.39 2.19 3.58	1.02 .75 1.34	1.39 1.02 1.82
8—KCl after N	{ A B Total	.49 1.09 1.58	.45 .21 .66	.25 .89 1.14	.36 1.03 1.39	.45 1.16 1.61	1.38 2.20 3.58	1.06 1.60 2.66	..... ..... .....	.33 .59 .92	1.02 .75 1.34	1.39 1.02 1.82
9—K <sub>2</sub> CO <sub>3</sub> after N	{ A B Total	.16 .50 .66	.12 .38 .26	.08 .30 .22	.03 .44 .47	.12 .57 .69	1.71 2.79 4.50	1.39 2.19 3.58	.33 .59 .92	..... ..... .....	1.02 .75 1.34	1.39 1.02 1.82

Differences in bold-face type are great enough for significance.

TABLE V-B  
DIFFERENCES BETWEEN TREATMENTS IN GRAMS

Treatment	Sample series	Treatments								
		1 A	2 A	3 A	4 B	5 B	6 A	7 A	8 B	9 B
CANE										
1—No K <sub>2</sub> O .....	A	....	.04	.24	.08	.05	<b>1.87</b>	<b>1.55</b>	<b>1.11</b>	.52
2—KCl with N.....	A	.04	....	.20	.04	.09	<b>1.83</b>	<b>1.51</b>	<b>1.07</b>	.48
3—K <sub>2</sub> CO <sub>3</sub> with N....	A	.24	.20	....	.16	.29	<b>1.63</b>	<b>1.31</b>	.87	.28
4—KCl after N.....	B	.08	.04	.16	....	.13	<b>1.79</b>	<b>1.47</b>	<b>1.03</b>	.44
5—K <sub>2</sub> CO <sub>3</sub> after N...	B	.05	.09	.29	.13	....	<b>1.74</b>	<b>1.60</b>	<b>1.16</b>	.57
NO VEGETATION										
6—KCl with N.....	A	<b>1.87</b>	<b>1.83</b>	<b>1.63</b>	<b>1.79</b>	<b>1.74</b>	....	.32	.76	<b>1.35</b>
7—K <sub>2</sub> CO <sub>3</sub> with N....	A	<b>1.55</b>	<b>1.51</b>	<b>1.31</b>	<b>1.47</b>	<b>1.60</b>	.32	....	.44	<b>1.03</b>
8—KCl after N.....	B	<b>1.11</b>	<b>1.07</b>	.87	<b>1.03</b>	<b>1.16</b>	.76	.44	....	.59
9—K <sub>2</sub> CO <sub>3</sub> after N...	B	.52	.48	.28	.44	.57	<b>1.35</b>	<b>1.03</b>	.59	....

Difference required: 19:1 = 1.01 grams

99:1 = 1.37 grams.

Differences in bold-face type are great enough for significance.

#### *No-Vegetation Treatments:*

During the first 45-day period, Treatment 6 lost an average of 1.38 grams more than Treatment 8, which was the no-vegetation treatment receiving no potash so far and which showed the highest average of potash leached. (Required difference for odds of 99:1 was 1.39 grams.) Compared with Treatment 1, the difference is 1.87 grams which represents 12.0 per cent of the potash applied. During the second period Treatment 6 lost 3.29 grams more than Treatment 1, or 21.1 per cent of the potash applied, making the total loss for the whole 90 days 33.1 per cent. This is undoubtedly a serious loss as compared with a 6.0 per cent loss of somewhat doubtful significance where cane was present.

Treatment 7 showed consistently smaller losses than 6, but the differences were not significant. Nor did Treatment 9 show a significant difference from Treatment 8, although the difference is in the direction to be expected from theoretical considerations. It is worthy of note that Treatment 8 showed significantly higher losses than the no-potash treatments, while Treatment 9 did not. In view of these considerations and of the fact that the same sort of thing was indicated on a smaller scale in the cane pots, there is strong indication that the carbonate was leached less than the chloride, but that the conservation of potash by its use was certainly not great enough to warrant applying potassium carbonate at a higher cost than the neutral potash salts. Could a source of carbonate of potash be found which would supply it at a cost comparable to or only slightly higher than the other potash fertilizers, its use would probably result in a small saving of potash.

Treatment 8B did not show significantly smaller losses than did 6A, but the difference was in the direction to be expected. Treatment 9B, however, showed a significantly smaller loss than 7A. Treatment 9B did not show significantly

lower losses than 1A and 1B (the difference required is 1.01 in both cases), but Treatment 7A showed very much greater losses than either 1A or 1B. In the case of Treatments 8B and 6A, much the same situation existed since the difference between 8B and 1A or 1B, while significant, is much smaller and much less reliable than that between 6A and 1A or 1B. Therefore, it seems possible to conclude that delaying the application of potash until after nitrification was completed resulted in lesser potash losses in the case of the no-vegetation pots.

In general it can be said that, except in the case of potassium carbonate applied after nitrogen, the no-vegetation pots receiving potash suffered very much greater losses of potash than did the cane pots. This demonstrates that utilization of applied potash by the cane crop is very rapid and efficient, surprisingly so, when it is remembered that the first potash application was only 14 days after the previous crop had been harvested. At this time there was very little aerial system developed by the crop. It is interesting to note that during the first period the no-vegetation pots which had received no potash (Treatments 8 and 9) lost very little more potash than did the cane pots (Treatments 1, 4 and 5), and that the amounts leached are all greater than the amount of potash (0.165 gram) which was originally present in the tap water which passed through the soil. This would indicate that a sort of equilibrium was set up with the water which had to be satisfied without regard to the presence or absence of cane. This does not imply that the cane was taking up no potash, since considerably more water had to be applied to the cane pots in order to keep their leachates comparable in volume with those from the no-vegetation pots and the additional potash which went on in this water was available to the plants.

It should be emphasized that the rapid utilization of potash herein indicated applies only to a ratoon crop where an extensive root system has already been established.\* This experiment does not enable prediction of potash losses from plant cane, but it can be assumed that they would be greater. In plant crops the use of nitrate fertilizers together with potassium carbonate until the root system had become extensive might result in considerable potash conservation. Such practice would probably be justified on acid soils with respect to potash conservation, provided the carbonate could be obtained at a price to compare favorably with the muriate or sulfate. This is demonstrated by the data from the no-vegetation pots which show that, except in the case of the carbonate applied after nitrification was complete, the applied potash was very inefficiently taken up by the exchange complex of the soil. It is evident that with acid soils the base-exchange complex can not be relied upon for retention of potash from neutral salts and that in the absence of an extensive root system serious losses of fertilizer may occur.

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\* Obviously, the conclusions herein suggested do not apply to acid soils which have been subjected to heavy rainfall within a few days after receiving applications of fertilizers containing potash. Under such conditions, leaching losses might be very great. It also is quite possible that, under similar circumstances, losses by leaching might be very considerable on a non-acid soil of reasonably high effective exchange capacity due to runoff of water from the surface and to channeling through the soil profile. Such losses would probably be as great with respect to applied nitrogen and might be considerable with respect to phosphate applications.

With respect to the problem of eventual serious damage to our soils from the use of acid-forming nitrogenous fertilizers through loss of effective exchange capacity and therefore ability to retain potash, these data indicate that while an acid soil is a poor retainer of potash, a ratoon crop of cane even shortly after harvest is a very good retainer of potash. Hence, it appears that the use of acid-forming fertilizers is unlikely to constitute a menace to our soils from this standpoint so long as they are cropped to cane. Nor, need the release of potash already present in the exchange complex of a soil resulting from the application of acid-forming fertilizer materials be considered a loss unless it results in luxury consumption, since it seems highly probable that it will be taken up by the crop. However, savings in potash applications might be effected with soils containing a good supply of exchange potash by making simple laboratory determinations of the amounts of potash released from such soils during nitrification, and decreasing the potash amendments to be applied by the amounts indicated. Such practice would tend to decrease potash losses due to luxury consumption by the cane.

In view of the fact that sugar cane thrives in acid soils and that, should the amounts of calcium, magnesium and the so-called "less essential" bases ever reach a dangerously low level due to use of acid-forming materials, they can be supplied economically in quantity necessary for plant nutrition, it appears that there is insufficient reason at present to discourage the use of these materials.

The demonstrably poor retention of applied potash by the exchange complex of an acid soil, together with the efficient uptake by the cane, indicates that applications of potash to such soils in quantities in excess of the crop needs will probably fail to build up the potash reserve of the soil and will result in loss due to luxury consumption by the crop. It seems likely that even in a non-acid soil the cane would tend to take up more of the excess than the soil and that such a policy would result in waste of fertilizer.

#### SUMMARY AND CONCLUSIONS

In ratoon cane pots, potash applied as either neutral salt or carbonate was not leached to any serious extent.

When potash was applied to cane pots at the same time as sulfate of ammonia was applied, significantly greater leaching of potash did not result than took place in pots which did not receive potash until nitrification was complete.

Somewhat lower losses of potash were indicated in uncropped pots which did not receive potash until after nitrification had taken place.

No important conservation of potash was gained by the use of the carbonate in cane pots.

Lower losses of potash were indicated by the use of the carbonate in uncropped pots.

Retention of applied potash by an acid soil in the absence of vegetation was poor, but in the presence of cane was very good, indicating highly efficient uptake of potash by ratoon cane.

This experiment indicated that seriously increased losses of potash by leaching as a result of long continued use of acid-forming fertilizers would be unlikely to occur in soils cropped to cane.



It was indicated that potash applied in excess of crop needs to an acid soil would probably fail to build up the reserve of exchange potash.

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## Disease Control and Stimulation of Cane Cuttings by the Hot-Water Treatment

By J. P. MARTIN and R. K. CONANT

Inasmuch as the hot-water treatment was first used experimentally in the Territory for the control of chlorotic streak disease and more recently as a plantation practice for stimulating the germination and early growth of cane cuttings, it seems advisable to present the subject matter in this paper in two parts. The first part which deals with disease control has been prepared by J. P. Martin while the second part which concerns a plantation practice has been written by R. K. Conant.

### DISEASE CONTROL BY THE HOT-WATER TREATMENT

The most effective control of any disease is the substitution of resistant varieties. The selection of healthy planting material is also highly efficacious for maintaining a low incidence of many diseases, especially in localities where environmental conditions favor their development and spread. Other of the more important measures of control are: roguing diseased plants, sterilization of cane knives, creating conditions favorable for cane growth, weed control, and more recently the hot-water treatment.

Sugar cane diseases such as gumming, leaf scald, and Sereh have been effectively controlled in other countries by the hot-water treatment. In experiments conducted here and elsewhere no control of mosaic has been secured by the treatment; no effect on the disease was evident even when diseased cuttings were immersed in water at temperatures from 52 degrees to 56 degrees C. for intervals from 10 minutes to 2 hours. At the higher temperatures and longer intervals cane growth was greatly depressed. However, at the lower temperatures and shorter intervals a stimulation in germination and cane growth resulted.

Chlorotic streak disease was first recognized in 1929 on POJ 36 at Olaa Sugar Company, Ltd., and since that time it has been found on all the Islands and on a large number of cane varieties. In pot studies pertaining to the disease at the Pathology plot it was established in October 1930 that symptoms of the disease failed to develop from cuttings of POJ 36 which were selected from diseased plants and given the hot-water treatment prior to planting.

In cooperative studies with R. K. Conant in relation to the field control of the disease, two small observational tests were installed in November 1930 at Olaa wherein diseased cuttings of POJ 36 were subjected to the hot-water treatment. Of the various temperatures, and intervals of exposures to such temperatures, the most satisfactory treatment proved to be 52 degrees C. for 20 minutes; in almost every instance this particular treatment has given 100 per cent control of the disease in cane cuttings. In these first experiments a definite growth response was apparent from the hot-water treatment (Fig. 1). Following the 1930 experiments many pot and field tests have been conducted on Oahu and Hawaii in relation to the cause and control of chlorotic streak and the response of varieties to the hot-water treat-



Fig. 1. Showing the effect of the hot-water treatment (52 degrees C. for 20 minutes) on cane cuttings of POJ 36 affected with chlorotic streak disease. Left, plants from healthy cuttings. Center, plants from diseased, treated cuttings. Right, plants from diseased, untreated cuttings. Note the superior growth from the diseased, treated cuttings (center).

—Photo by Conant.



Fig. 2. Chlorotic streak disease is controlled in cane cuttings by the hot-water treatment. Left, cane growth resulting from diseased cuttings of 31-2806 following the hot-water treatment. Right, cane growth from diseased, untreated cuttings of 31-2806.—Photo by Conant.

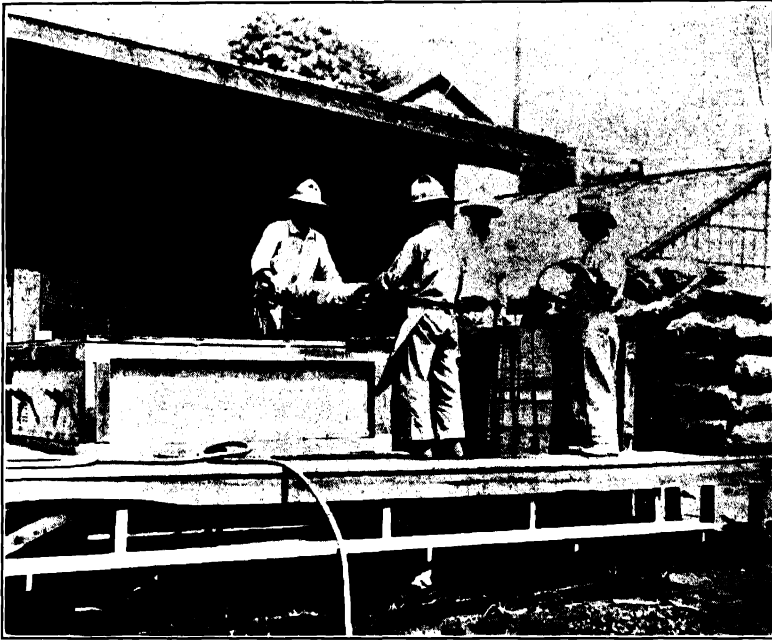


Fig. 3. Hot-water unit for treating cane cuttings installed in 1931 at Makiki.

ment, the results of which have demonstrated that the disease, in so far as it is carried by cuttings, is controlled by the treatment (Fig. 2), and that the germination and early growth of most varieties are stimulated. It should be stated that with a few varieties the treatment resulted in depressed growth.

Following the appearance of chlorotic streak disease on canes at Kailua substation, it was deemed advisable to subject all cuttings to the hot-water treatment prior to shipment to plantations and regional stations; in order to carry out this procedure a hot-water unit (Fig. 3) was constructed in 1931 at the Makiki station. The water in the large tank is heated and maintained at 52 degrees C. by steam, and to insure a uniform temperature the water is circulated by a centrifugal pump. Immediately following the treatment, the cuttings are placed in a tank of cold water, as shown in the extreme left of Fig. 3, to prevent possible injury from prolonged heating. Since it was later found to be more economical to treat and pack cane material for shipment at Kailua substation, a hot-water unit was installed there in 1934.

The tanks first used for treating large quantities of cane cuttings were constructed with redwood but later it was found that large metal tanks could be used to advantage. The size of the tank depends on the quantity of material to be treated; some of the first tanks were 4 feet wide, 6 feet long, and 5 feet deep, while the metal tanks now in use at Waipio and Kailua substations are somewhat larger.

After considering the advantages of the hot-water treatment in relation to additional ways and means for excluding foreign cane diseases and insects with imported cane material, the Quarantine Committee in 1931 adopted the practice of subjecting to the hot-water treatment all cuttings of imported cane varieties upon their arrival

into the Territory and all cuttings of varieties released from the Molokai Quarantine Station.

In 1931, C. E. Pemberton demonstrated that all observed insects, either within or on cane cuttings were killed at an exposure of 52 degrees C. for 20 minutes. In the Director's Monthly Report for August 1933, Mr. Pemberton states: "In cooperation with the Agriculture department, a test was completed to determine a satisfactory hot-water treatment for cane cuttings shipped as seed from Anomala territory to plantations outside its known distribution. Living *Anomala* adult beetles were bagged with cane seed having dry cane leaves for packing and the entire package immersed in water at 50 degrees C. for 30 minutes. No beetles survived this treatment and we understand this degree of heat for 30 minutes is not harmful to the cane seed. In the future, this will be the standard treatment for all cane seed shipped from Anomala infested fields to other localities in Hawaii." Since that date (1933), when a hot-water unit was completed at Waipio, all cane material shipped from Waipio has received such treatment (50 degrees C. for 30 minutes).

In recent years Ceresan, an organic mercury compound, has been found by staff members of plantations and this Station to be the most effective disinfectant for treating the freshly cut ends of sugar cane cuttings for preventing the entrance of organisms which, under wet and cold climatic conditions, often cause a rotting and souring of the cuttings. The cut ends are dipped in a one per cent (by weight) solution of Ceresan. The directions regarding the application of Ceresan and its effectiveness are summarized in a circular letter by the Genetics department dated February 27, 1939. Ceresan apparently does not stimulate germination *per se* but a superior growth results since it prevents the entrance of rot-producing organisms which play a major part in poor germination.

#### HOT-WATER TREATMENT OF CANE SEED

The stimulating effect on germination, produced by treating cane seed in various ways prior to planting, has often been discussed in the literature. Soaking cane seed in cold water from 24 to 48 hours is an old method of treating cane seed and has been used successfully by plantations in the Hawaiian Islands for many years. The addition of lime to the cold water is said to further hasten germination of the seed. Soaking cane seed for one hour in water treated with acetylene gas has also been found to stimulate germination of certain varieties. More recently a method in which the freshly cut ends of seed pieces are dipped into a one per cent Ceresan solution has been used quite widely. The senior author of this paper found that treating cane seed in hot water at 52 degrees C. for 20 minutes resulted favorably in controlling chlorotic streak disease. He also found that this treatment hastened germination and stimulated growth. These methods of treating cane seed have been tried experimentally at Olaa Sugar Company, and the cold- and hot-water treatments have been used on seed planted to field-scale proportions.

The results of experimental work in cooperation with Mr. Martin on the chlorotic streak disease problem at Olaa Sugar Company left no doubt as to the value of treating seed with hot water under conditions on this plantation, not only in relation to chlorotic streak disease, but also as a practical means of hastening germination, stimulating growth, and reducing cultivation costs in plant cane. Therefore,

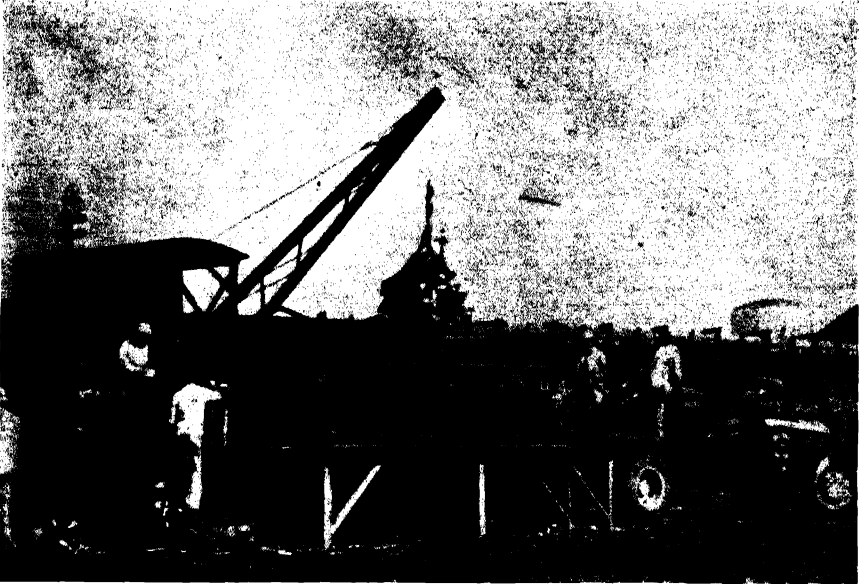


Fig. 4. Hot-water unit used at Olaa Sugar Company for treating cane cuttings. The tank is 12 feet x 12 feet x 6 feet deep with a capacity of 6463 gallons. The tank can handle 100 bags of seed at a time. The small, inexpensive loading machine is operated by a 6-horsepower, one-cylinder gasoline motor. This unit is able to treat 2000 bags per day with 4 men assisting in the work at a cost of less than one cent per bag. Additional transportation costs involved in trucking the seed to and from the unit brings the total cost for hot-water treating of seed to about one cent per bag.—*Photo by Conant.*

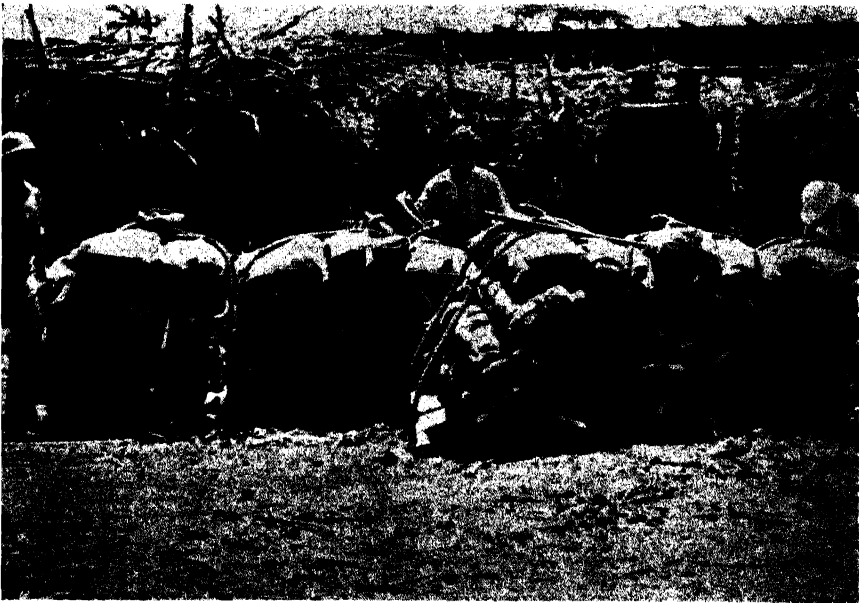


Fig. 5. Showing the method used at Olaa Sugar Company for piling the bags into rope slings preparatory to handling with the small loading machine shown in Fig. 4.—*Photo by Conant.*

this method of treating cane seed was developed to a point where large quantities of seed can now be handled with ease, and today all seed at Olaa Sugar Company is given this treatment prior to planting. The unit shown in Fig. 4 was built to handle the hot-water treating of seed, and has proven very satisfactory. This unit can handle 2000 bags per day, but during the planting season so far this year 1000 to 1500 bags per day have been sufficient to take care of the planting requirements. Four men working 8 hours a day can attend to piling 1000 bags into slings as shown in Fig. 5 and assist in the work in general. A careful check is kept on the temperature of the water which is not allowed to vary more than 2 degrees (50 degrees C. to 52 degrees C.). The water is heated by means of steam which enters through a perforated 1-inch pipe at the bottom of the tank. When the maximum temperature of 52 degrees C. is attained, the steam is shut off and about 100 bags of seed are lifted into the tank, 10 bags at a time in slings as shown in Fig. 4. The temperature is maintained by allowing steam to enter slowly during the 20 minutes the bags are being treated.

When it is convenient to do so the bags are piled in slings on the trucks that transport the seed to the hot-water unit, thus saving one step in the handling of the seed.

Experiments at Olaa Sugar Company have indicated that it is unnecessary to dip seed in cold water following the hot-water treatment, and that the temperature may vary between 50 degrees C. and 52 degrees C. and the time between 20 and 25 minutes without altering the results. These details may seem of minor importance, but they have a bearing on the method when considering treatment of seed on a large scale. If it is permissible to use the leeway in time and temperature mentioned, and plant the seed without dipping in cold water following the hot-water treatment, then the method becomes much easier to adopt for practical purposes.

The hot-water treatment has a tendency to soften the eyes and unless precautions are taken to prevent mechanical injury to the eyes, poor stands may result from hot-water treated seed. This applies particularly to body seed, since the eyes of body seed, unlike the eyes of top seed, are not protected from mechanical injury by the leaf sheath. Various ways of handling body seed have been tried at Olaa Sugar Company with indications that the simplest and most effective method is to insist that the seed cutters leave the trash on the stalks and seed pieces, and when bagging the seed to place a little trash in the bags with the seed. The trash protects the eyes from mechanical injury.

During the first 6 months of 1939, 60,000 bags of seed have been hot-water treated at Olaa Sugar Company at a cost of about one cent per bag. From previous experience we are quite certain that this small additional planting cost will be more than offset by reduced cultivation costs for weed control, brought about by the stimulation in growth due to the treatment.

A comparison between 1 per cent Ceresan-treated and hot-water treated seed is shown in Fig. 6. The variety in this test is 31-2484 planted 21 days before the photograph was taken. The test is located in a mauka field, elevation 1650 feet, where conditions are everything but favorable for rapid germination. The seed planted in the C plot on the right was treated with a 1 per cent Ceresan solution in the usual manner by dipping the freshly cut ends of the seed pieces into the solution.

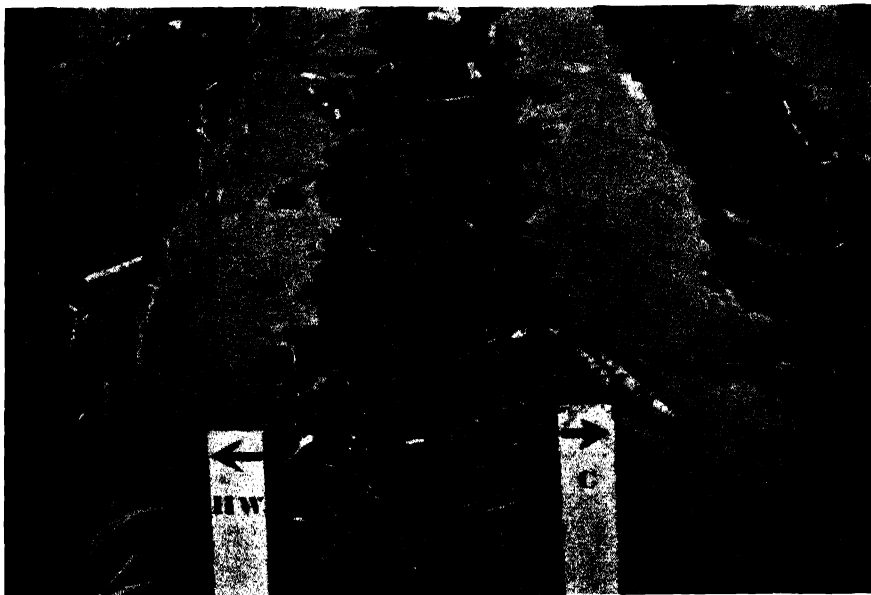


Fig. 6. Showing a comparison between hot-water treated seed (HW) at the left and Ceresan-treated seed (C) at the right. Photograph taken 21 days after the seed was planted. Cane variety 31-2484, elevation 1650 feet. The cane shoots in the HW plot can be easily seen, whereas the shoots in the C plot do not show up in the photograph since they were only commencing to penetrate the mulch paper at the time the photograph was taken. A plot which adjoins the C plot, but not shown in the photograph, was planted with untreated seed, and after 31 days no signs of germination could be seen in this plot. No signs of germination could be seen in this untreated plot until 5 weeks after planting.—*Photo by Conant.*

The seed in the HW plot on the left which was planted on the same day, was given the hot-water treatment, viz., soaked in hot water at 52 degrees C. for 20 minutes. A count was made in this test of the number of shoots that had germinated and penetrated the mulch paper in 21 days. The data are given below :

Plot	No. ft. of row	No. shoots	Plot	No. ft. of row	No. shoots
HW	100	73	C	100	35
HW	100	73	C	100	17
HW	100	78	C	100	6
HW	100	106	C	100	2
HW	100	99	C	100	13
Total		429	Total		73
Average		86	Average		15

The average height of the shoots in the HW plot 21 days after planting was 12 inches against 3 inches for the shoots in the C plot.

A check plot not included in the photograph was planted with untreated seed on the same day. This plot adjoins the C plot and after 21 days no signs of germination could be seen. All plots in this test are  $\frac{1}{2}$  acre in size, and were planted for observational purposes.

The plant fields at Olaa Sugar Company are doing exceptionally well this year despite the unusually cold and wet weather that has prevailed and slow and poor



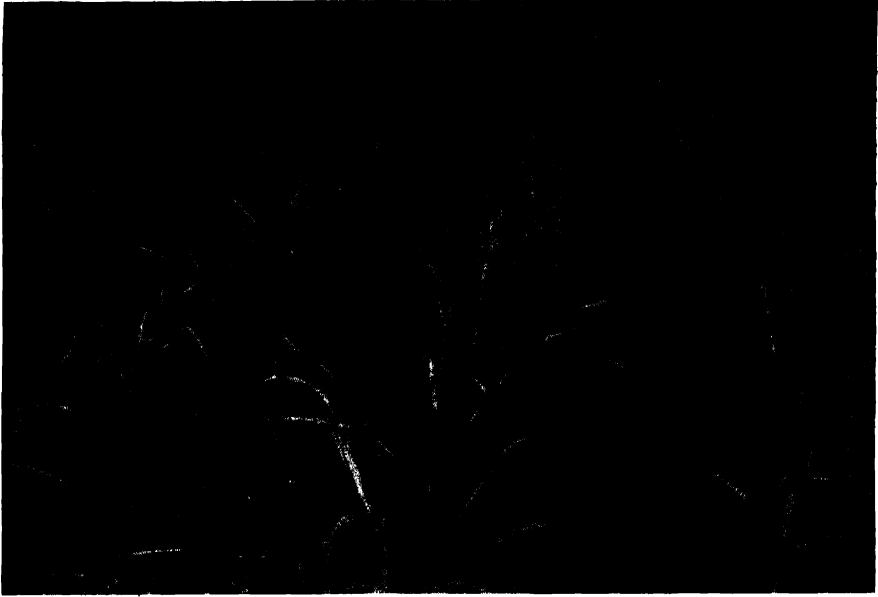


Fig. 7. Showing portion of a field of 32-1063, 6 weeks after planting. Note the full, vigorous stand obtained from hot-water treated seed. Elevation of this area is 1650 feet where annual rainfall usually exceeds 200 inches, and average maximum temperatures rarely exceed 73 degrees F. and average minimum temperatures are seldom above 61 degrees F.

—Photo by Conant.

germinating canes have germinated rapidly with full stands even when planted at high elevations. Fig. 7 shows 32-1063 plant at 1650 feet elevation 6 weeks after having been hot-water treated and planted.

#### *Discussion:*

At present the stimulating effect from the treatment is not understood. However, it is believed to stimulate certain chemical and enzymic changes within the cuttings which are normally associated with germination. Another theory is that the treatment may increase the rate of cell division of the meristematic cells in the lateral buds and accelerate those physiological processes (absorption, respiration, etc.) accompanying growth.

It is interesting that the hot-water treatment should be so effective for controlling chlorotic streak disease and at the same time for stimulating the germination and early growth of sugar cane cuttings.

Where chlorotic streak is a problem or when questionable planting material is to be used, it is recommended that such material be treated in order to secure healthy planting material. As an aid for better germination, especially at the higher elevations where environmental factors are less favorable for cane growth, it is hoped that the heat treatment will be used or at least be experimentally tested with different varieties under various conditions.

A growth response immediately following planting, as already pointed out, reduces weeding and cultivating costs and insures a more uniform stand of cane. The

sooner a cane plant establishes its own shoot roots and becomes independent of the original cutting, the better chance it has for competing against adverse environmental factors and for maintaining a normal growth. An uneven growth of plant cane under cold and wet conditions has been caused at times by rot-producing organisms attacking cane cuttings prior to and during germination; the use of Ceresan has aided greatly in protecting cuttings from such attacks. Any treatment that will hasten the germination of sugar cane cuttings will make for better growth and lower field costs.

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## Evaporation of Moisture From Soil in Large Lysimeter Pots

By P. L. Gow

While conducting a lysimeter experiment on the leaching of potash from a soil, it was found necessary to determine the daily water consumption of the pots used in the experiment. The water consumption was calculated as the difference between amounts of water applied to each pot by irrigation and rainfall, and amounts of water collected as drainage. The experiment required the inclusion of 16 lysimeter pots which were kept free of all vegetation. Since the water consumption of these no-vegetation pots seemed to indicate that considerable quantities of water were removed from the soil by evaporation, contrary to certain established ideas with respect to soil-water relationships, these data were deemed worthy of reporting.

The general experimental layout is described elsewhere.\* The lysimeters consisted of cubical concrete pots 2 x 2 x 2 feet, which had received three coats of hot asphaltum to prevent leakage. The entire layout consisted of 36 pots arranged in 4 rows of 9 pots each. The pots were buried to ground level and between each pair of rows was a sunken concrete runway which provided for collection of drainage. The drainage from each pot was received in an especially constructed 5-gallon tin can, the inlet of which had a diameter of 1.5 inches so that evaporation from the cans was minimized. The soil used was a free-draining, acid Manoa soil, which had been air-dried, screened and well mixed before placing in the pots. Pots containing cane alternated with no-vegetation pots.

Water received as rainfall was measured by means of a standard rain gauge. Water applied as irrigation was weighed on a solution balance sensitive to 1 gram. Drainage was measured by weighing the cans which contained water, emptying and re-weighing the empty cans for tare. One gram of water was considered as equivalent to 1 c.c. and water data were recorded as cubic centimeters.

All pots were fertilized alike with respect to nitrogen and phosphorus. Potash fertilization was varied.\* All pots had contained sugar cane which was allowed to develop until several nodes of millable stalk showed above ground. The cane was then cut back to the seed piece in the no-vegetation pots so as to remove all points from which new growth might start. The no-vegetation pots were kept free of weeds throughout the time that drainage was collected.

Immediately after the cane had been cut and before the collection of drainage data reported in Table II had begun, it was noticed that the water consumption from all pots was very appreciable. To test whether this was due to evaporation, 12 of the 36 pots were covered with crudely fitted cardboard covers. The data presented in Table I show the water consumption of each pot during the 24-hour period following. The results from this rough experiment are taken to indicate

\* Gow, P. L., 1939. A Lysimeter Study of Losses of Applied Potash by Leaching from an Acid Soil. *The Hawaiian Planters' Record*, 43: 263-276.

that the water consumption was chiefly due to evaporation. It is probable that the water which disappeared from the covered pots was also evaporated, since the covers were by no means tight.

TABLE I  
COMPARISON OF WATER CONSUMPTION OF COVERED AND  
UNCOVERED POTS

Covered	Uncovered	
c.c.	c.c.	c.c.
1078	2846	2524
1059	2700	2491
783	2704	2106
626	2518	2359
1103	2561	2503
853	2529	2341
723	2919	2150
813	2802	2149
953	2507	2246
772	2352	2411
735	2503	2261
562	2093	2188

Table II presents the average water consumption data from the no-vegetation pots. Drainage measurements were made the morning of the day following irrigation, the latter having been applied in the late afternoon so that the pots would still be draining at sundown. Thus, daily consumption data are calculated on the assumption that each pot had a definite water-holding capacity and that evaporation reduced the water held below this level. The difference between water applied in the afternoon and water drained next morning was taken as water consumed the previous day.

Occasionally, the drainage pipes of one or more pots became stopped and were not freed until next day. These stoppages were always indicated in the drainage data, and drainage figures from such pots were discarded in calculating the average consumption figures until it was certain that normal drainage had been re-established. Thus, the data presented contain no values known to be abnormal due to stoppage of drains. Twice, due to heavy rainfall, and once, due to some unknown cause, drainage data from all pots were considered unreliable, so no data for the periods August 3-4, August 9-10 and August 24 have been reported.

TABLE II

Date	Average total c.c.	Average total inches	Average daily inches	Date	Average total c.c.	Average total inches	Average daily inches
1938				August			
June				8	2369	0.25	0.25
25-26	4500	0.48	0.24	9-10	....	....	....
27	3630	0.38	0.38	11-15	10799	1.14	0.23
28	3199	0.34	0.34	16	3918	0.41	0.41
29	2227	0.24	0.24	17-19	2154	0.23	0.08
30	1770	0.19	0.19	20-21	4375	0.46	0.23
July				22	2404	0.25	0.25
1-4	6719	0.71	0.18	23	2577	0.27	0.27
5	2740	0.29	0.29	24	....	....	....
6	2620	0.28	0.28	25	952	0.10	0.10
7	2977	0.32	0.32	26-28	7580	0.80	0.27
8	3055	0.32	0.32	29-30	4516	0.48	0.24
9-10	2847	0.30	0.15	September			
11	2247	0.24	0.24	31-1-2	7581	0.80	0.27
12	1996	0.21	0.21	3-5	6447	0.68	0.23
13	2972	0.31	0.31	6	3373	0.36	0.36
14	3274	0.35	0.35	7	1240	0.13	0.13
15	2942	0.31	0.31	8	2465	0.26	0.26
16-17	4310	0.46	0.23	9	2845	0.30	0.30
18	3034	0.32	0.32	10-11	2173	0.23	0.12
19	2461	0.26	0.26	12	2198	0.23	0.23
20	4114	0.44	0.44	13	2452	0.26	0.26
21	2297	0.24	0.24	14	2151	0.23	0.23
22	1972	0.21	0.21	15	1704	0.19	0.19
23-24	4878	0.52	0.26	16	2368	0.25	0.25
25	3946	0.42	0.42	17-19	6173	0.65	0.22
26	2738	0.29	0.29	20-21	5166	0.55	0.27
27	3251	0.34	0.34	22	2730	0.29	0.29
28	4413	0.47	0.47	23	1831	0.19	0.19
29	4452	0.47	0.47	24-25	4645	0.49	0.25
30-31	3367	0.36	0.18	26	1386	0.15	0.15
August				27-28	3643	0.39	0.19
1	2132	0.23	0.23	29	2163	0.23	0.23
2	2849	0.30	0.30	October			
3-4	....	....	....	30-1-2	4833	0.52	0.17
5	2451	0.26	0.26	3-4	4694	0.50	0.25
6-7	4363	0.46	0.23	5-6	4341	0.46	0.23

Table III presents average total consumption figures for successive 30-day periods.

TABLE III

Period	No. of days	Average total consumption, inches
1938		
June 25-July 24.....	30	7.72
July 25-August 23.....	30	6.61*
August 24-September 22.....	30	6.79*
September 23-October 6.....	14	2.93
	<hr/> 104	<hr/> 24.05

\* Data for 4 days in the period July 25-Aug. 23 and for 1 day in the period Aug. 24-Sept. 22 are missing. If these missing figures are estimated as equivalent to the average daily consumption for the periods, the totals become 7.63 inches and 7.02 inches, respectively. The total for the entire 104 days becomes on this basis 25.30 inches. This procedure is probably not justified in the period July 25-Aug. 23 since the missing figures are due to heavy rainfall, during which evaporation would be expected to be low.

#### DISCUSSION OF DATA

Since the data presented in Table I appear to show that water consumption of these no-vegetative pots was due to evaporation, consumption data will be considered as evaporation hereinafter. Most of the days during this experiment were hot and sunny. On such days, the surfaces of the pots became completely dry to a depth of at least one-half inch by 10:00 A.M. with the exception of one pot, the surface of which for some unknown reason never became dry. This pot drained well and its consumption data showed no significant difference from that of the other 15 pots. Evidently this was a case in which water moved upward in the soil due to capillary action. In the case of the other pots, most of the water lost by evaporation must have moved up from the lower soil levels as water vapor since the surface soil was dry most of the time evaporation took place.

That the high rate of evaporation showed by these pots was not entirely due to the fact that they were usually irrigated daily is demonstrated by the data for periods when the pots received no irrigation for several days. Average daily evaporation in inches for July 1-4 was 0.18 inch. During the period only 0.02 inch of rain fell. During the period August 11-15, the average daily evaporation was 0.23 inch. The rainfall was: August 11, 0.12 inch; August 13, 0.11 inch, and August 15, 0.02 inch. The data from these and other periods when the pots did not receive daily irrigation indicate that, while the average daily evaporation tended to be somewhat lower during these periods, it was comparable with the evaporation when the pots were irrigated daily.

The author is not qualified to draw any general conclusions concerning soil-water relationships from these data. They were secured during the progress of an experiment designed entirely for other purposes and are presented here only in the hope that they may be of interest to workers in the field of soil irrigation.

## Sugar Prices

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96° CENTRIFUGALS FOR THE PERIOD  
JUNE 21, 1939 TO SEPTEMBER 15, 1939

Date	Per pound	Per ton	Remarks
June 21, 1939.....	2.81¢	\$56.20	Philippines, 2.80, 2.82.
“ 22.....	2.85	57.00	Philippines.
“ 27.....	2.87	57.40	Philippines, 2.85; Puerto Ricos, 2.86, 2.90; Cubas, 2.90.
“ 28.....	2.90	58.00	Philippines, Puerto Ricos.
July 5.....	2.93	58.60	Puerto Ricos, Philippines.
“ 11.....	2.85	57.00	Puerto Ricos.
“ 14.....	2.84	56.80	Philippines.
“ 20.....	2.90	58.00	Cubas.
“ 21.....	2.88	57.60	Philippines.
“ 24.....	2.90	58.00	Puerto Ricos.
“ 25.....	2.92	58.40	Cubas.
“ 26.....	2.90	58.00	Philippines.
Aug. 7.....	2.85	57.00	Philippines.
“ 8.....	2.82	56.40	Cubas.
“ 9.....	2.80	56.00	Cubas.
“ 15.....	2.82	56.40	Philippines.
“ 16.....	2.83	56.60	Philippines.
“ 22.....	2.80	56.00	Puerto Ricos.
“ 25.....	2.90	58.00	Philippines.
“ 31.....	2.92	58.40	Puerto Ricos.
Sept. 5.....	3.75	75.00	Philippines.
“ 6.....	3.81	76.20	Cubas, 3.76, 3.81, 3.86.
“ 7.....	3.80	76.00	Cubas, 3.79, 3.81.
“ 8.....	3.85	77.00	Philippines.
“ 13.....	3.60	72.00	Philippines.
“ 14.....	3.70	74.00	Philippines.
“ 15.....	3.60	72.00	Philippines.

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